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ON SOME PRACTICAL APPLICATIONS OF IMPREGNATED, DRIED PAPER DISCS CONTAINING VARIOUS REAGENTS FOR THE CULTURAL DIFFERENTIATION OF BACTERIA.*

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The present paper supplements the preliminary description by Snyder[1] of a new and simplified method for the cultural differentiation of bacteria. In brief, it consists of the drying of paper discs previously moistened with various reagents. These may be stored and placed as needed on inoculated agar media in Petri dishes. Multiple tests can be performed with a minimum of effort and materials. At least three possible reactions occur: sharp zones of diffusion around the discs; color changes in the pads themselves, or reactions which may be developed on the pads by chemicals. This method has many practical applications in the fields of teaching and diagnositic bacteriology. Its greatest value would appear to lie in the fact that these discs can be impregnated with not only the common carbohydrates and chemicals used for differential tests but also the entire culture medium. The former requires only a nutrient agar for a base, the latter nothing but a salt-agar mixture. The resultant simplification of media making and reduction of storage space should be of interest to all laboratories.

The economy and practical value of dried paper discs containing carbohydrates for fermentation studies was recently reported by Soto[2] but he put the discs in separate tubes and added small amounts of young peptone broth cultures. Other than keeping

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the sugars dry and in a reduced space, he made no particular advance over the employment of a drop of concentrated sugar in a tube of basic medium. Our method is adapted from one described by Knox[3] for the preliminary screening of intestinal bacteria. He heavily streaked the surfaces of two separate media in a Petri dish with the suspected colony, after which strips of filter paper moistened with mannite and sucrose were placed over the path left by the inoculating wire. A coverslip adjacent to the mannite trapped the bubbles of gas when the latter was fermented and another coverslip aided in the detection of hydrogen sulfide. He did not mention the use of possibilities of dried filter paper strips for specific cultural differentiation. His procedure with its two media and coverslips seemed too cumbersome and expensive for widespread application. An unsuccessful attempt to apply this method to another problem led to a modification employing heavily inoculated "poured" plates. The striking reactions in the medium which contained phenol red as the indicator suggested the technique to be discussed.

MATERIALS

1. THE PAPER PAD: For our purposes bioassay discs (Schleicher and Schuell #740 E) of ½ and ⅜ inch diameter were found to be ideal. The ¼ inch disc increased the number of tests possible in a Petri dish, but it did not carry enough load of carbohydrate to prevent rapid exhaustion with subsequent reversion from an acid to an alkaline reaction. The discs were first dry heat sterilized and then moistened with sterile or freshly prepared solutions, after which they were dried 15-20 minutes in the open under an infra-red lamp. This time and temperature prevented "toasting." These dried pads were marked with india ink for identification and stored in any satisfactory receptacle.

2. THE REAGENTS: The tests were first limited to discs impregnated with the usual differential sugars, urea and lead acetate. The carbohydrates were in 50 per cent solutions but later reduced to 20 per cent when used alone with the nutrient base. About 8 of the ½-inch discs could be prepared with 1.0 ml. of these solutions. These dried discs were, in the beginning placed on a sugar-free medium containing an indicator, but the practical problem of trying to develop reactions with similar colors to the indicator made it imperative to put the indicator in the pad. Considerable difficulty was encountered in finding a suitable indicator for the discs. The phenol-sufonphthalein dyes, brom-cresol-purple, brom-thymol-blue, and phenol-red, had the objectionable feature of turning yellow on the acid side and thereby were hard to distinguish in zones of acid in a straw-colored or translucent medium. Furthermore, the developing

bacterial growth left the pads colorless. This required an additional drop of indicator to be put on them at the time of reading. It was only when the medium reverted to the alkaline side that the colors could be easily observed. Likewise, the diffusion of the indicators frequently interfered with the development of reactions by chemicals on neighboring pads, such as the detection of acetyl-methyl-carbinol (the Voges-Proskauer test) with concentrated sodium hydroxide. For these reasons, neutral red was chosen since it was red on the acid side and less diffusible than the other indicators. It was added in approximately 1-20,000 concentration and the mixtures made alkaline to color with 1 N sodium hydroxide.

As the method was studied, it was found possible to incorporate the entire culture medium in the disc. This required the sugar-free nutrient base to be in the form of a 10 per cent solution to which the carbohydrates were added in 40 per cent or saturation quantities to assure the desired reactions. This step accomplished two things: the reduction of the base medium to one of salt and agar, and secondly, the control of the nutritional requirements as in the CITRATE test in the IMVIC reactions where the source and nature of the carbon must be known. Actually, this can be said to approach the ideal, since it is possible to develop any or all reactions on a non-nutritional base and not be bothered by the complexities of large-scale nutrient medium manufacture. This procedure can be said to be a modification of the "auxanogram" technique described over a half-century ago by Beijerinck.[4] The discs impregnated with these concentrated solutions must be handled carefully in the drying process since objectionable browning or "toasting" quickly occurs. It should also be noted that the concentrated solutions used for impregnation cannot be autoclaved because they darken beyond possibility of use, but they can be filtered while hot.

3. THE MEDIA: As previously mentioned, the media necessary for this method have been reduced to two: a nutrient agar and a salt-agar base. Several combinations of peptones, meat extract and/or casein digests have been tested to provide a satisfactory sugar-free base. Of these ingredients, a medium consisting of 2 per cent tryptose (Difco), 0.5 per cent sodium chloride and 1.2 per cent agar has given excellent results. This amount of agar provides sufficient moisture yet prevents too much diffusion. The salt-agar mixture was composed of 1.2 per cent agar and 0.5 per cent sodium chloride. The media was adjusted to pH 7.2, tubed in 15-20 ml. amounts and autoclaved at 120 C for 15 minutes.

METHODS AND RESULTS

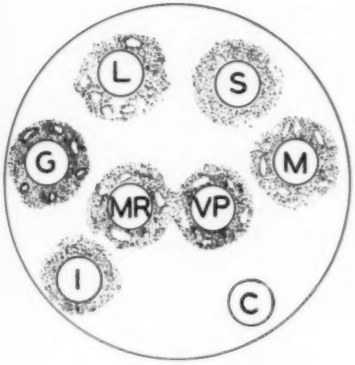
The method is very simple. Tubes of media are melted in boiling water, cooled to 50 C and inoculated with varying amounts of aqueous suspensions of young broth cultures of the organisms being studied. "Poured" plates are made. After solidification of the agar, the discs, each containing the desired reagent, are distributed over the surfaces of the medium in the Petri dishes. The plates are incubated at 37 C and examined in 3-36 hours dependent upon the size of the inoculum, the organism, and reactions or end-products desired.

The reactions which occur are interesting and usually easy to read. This indicator, neutral red, turns red in the presence of acid caused by fermentation of carbohydrates; gas bubbles are formed in the zone of diffusion around the disc by those species capable of this reaction. Hydrogen sulfide blackens the paper containing lead acetate. Indol can be determined by a drop of Kovacs' reagent placed on the pad. The Voges-Proskauer test can be quickly developed by O'Meara's creatine-sodium hydroxide method. These are but a few of the tests which can be performed with this technique. The differentiation of some of the organisms of the so-called "C-T-D" group is shown in the accompanying illustrations.

A word should be said about the rate of the reactions. In respect to the organisms of the above group, the majority of tests needed for their differentiation can be determined within three hours by means of large inocula. We have been able to demonstrate the production of acid and gas, indol and hydrogen sulfide, and hydrolysis of urea within this period by means of 1.0 ml. inocula of young broth cultures or moderately light aquesous suspensions of young cultures. The time in which these end-products appear can be appreciably slowed by use of the conventional loop. Thus, some degree of flexibility to meet the needs of time can be provided by this method. It also creates problems of reactions occurring too quickly for student work in laboratory periods separated by a day. The staphylococci and streptococci develop at a slower rate for the most part than the intestinal forms.

Until recently, the "poured" plate was used entirely in order to detect gas formation because paper pads do not apparently entrap gas bubbles as the coverslips do in the technique of Knox.[2] Soto[1] also noted that gas was not detectable in tubes containing the paper disc and broth culture. He suggested that the presence of gas in such preliminary media as Kligler's Iron agar was indicative of such formation in other carbohydrates and hence, made it necessary only to know the actual formation of acid. This modification can be applied to our method when the

FIG.1 TYPICAL CULTURAL REACTIONS WITH IMPREGNATED PAPER DISCS.



GLUCOSE	AG
LACTOSE	AG
SUCROSE	O
MANNITE	AG
INDOL	+
M.R.	+
V.P.	-
CITRATE	-

A. STRAIN OF ESCH. COLI IN AGAR BASE

GLUCOSE	A
LACTOSE	O
MALTOSE	A
MANNITE	A
INDOL	+
H_2S	-
UREA	-

B. STRAIN OF SH. PARADYSENTERIAE IN NUTRIENT AGAR.

organisms are spread over the surface of the medium. This is a particularly applicable approach in the case of organisms which do not form gas or so few tests are required that more than one series can be performed on a single plate as in the IMVIC tests proper.

To date the method has been tested with a large group of "coliform" organisms and stock culture of intestinal pathogens. Consistent and reliable results were obtained for the production of acid from carbohydrates, formation of indol, hydrogen-sulfide and acetyl-methyl-carbinol, and the utilization of citrate compared with similar reaction occurring in test tubes inoculated simultaneously with the plates. However, gas formation was not particularly reliable compared with that found in the conventional Dunham tube, and it would appear that this detection will always be a problem with those species producing only small amounts or traces of gas. Hydrolysis of urea was obscured by the rapid rise in the pH of the medium in the zone of growth as the organisms metabolized and the indicator, phenol red, was turned pink. Similarly, this accumulation of metabolic products in the relatively thin layer of medium frequently caused a reversion to the alkaline side of the indicator within 12-18 hours, a feature which was confusing unless gas, a reliable indicator of fermentation, was formed. None of these objections, except possibly gas formation, seems to present an unsurmountable obstacle for solution. Despite these shortcomings, it is still felt that this method already offers a definite improvement over present methods for the cultural differentiation of bacteria.

DISCUSSION

The authors do not know of a method where paper discs, pads or strips have been employed to yield so much information with the amount of medium contained in a Petri dish, although paper has long been used in the evaluation of disinfectants and antibiotics as well as detection of hydrogen sulfide and indol. The present technique offers distinct advantages over the rapid methods advocated by Weaver and his associates[5,6] who conducted them in separate tubes, and also retains the desirable feature of reactions limited to a single reagent or sugar in contrast to the double or triple sugars and similar types of media. The greatest asset of the method is that the pad with its constituents may be dried and stored for future use. They can easily be prepared in large numbers, identified in a number of ways for ready recognition, and kept in a space much smaller than that for the amount of media necessary to do an equivalent number of tests in tubes.

This technique should also have great value in the teaching of bacteriology; first, a majority of the differential tests com-

monly taught can be carried out in a single Petri dish in contrast to a series of test tubes all too frequently confusing to the student, and second, the impregnated discs for an entire course could early be distributed as desk equipment to be used when needed. Thus, most of the requirements for each organism would be a Petri dish, a tube of basic medium, and storage facilities. it should offer a way to eliminate some of the multitudinous types of culture media employed for the cultural differentiation of bacteria. This feature should also appeal to the small hospital or private laboratory where the frequent spoilage of tubes of media due to drying or contamination results in higher costs for each organism than would occur with an equivalent number of tests using these discs.

In closing, however, it should be stated that the method is still in an experimental stage and that no claim is made at this time for completeness or accuracy beyond that described. It is offered as a possible solution for many cultural problems, but much more work is needed to establish its place in Bacteriology.

SUMMARY

A method employing dried paper discs containing various reagents has been developed for the cultural differentiation of bacteria. Its advantages in respect to simplification of media requirements and storage facilities with concomitant accuracy of results for certain reactions have been described.

BIBLIOGRAPHY

1. Snyder, M. L. Science, In Press.
2. Soto, O. B. Puerto Rico J. Pub. Health. 25: 96, 1949.
3. Knox, R. J. Path. and Bact. 61: 343, 1949.
4. Beijerinck, M. W. Arch. Neelanderlaises. 23: 367, 1880.
5. Weaver, R. H., Arnold, W. M. and Hannan, J. J. Bact., 51: 565, 1946.
6. Arnold, W. M., and Weaver, R. H. J. Lab. and Clin. Med. 33: 1338, 1948.

NEWER METHODS FOR THE DETECTION OF Rh ANTIBODIES

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The study of the Rh antigens and antibodies and the techniques of testing have developed with amazing speed since the original discoveries by Levine and Stetson,[12] Landsteiner and Wiener.[11] Due to the importance of the Rh problem to child birth and transfusion, the determinations of Rh antigens and antibodies have become essential serological procedures for the clinical laboratory.

The technologist must keep in mind some of the serological and chemical properties of the antibodies involved, in order to be sure that the antibody detection technique fulfills the purpose for which it is employed. These antibodies fall into three categories according to their serological and chemical behavior[6, 7, 9] as seen in Figure 1. Not all methods are usable for their detection. Some techniques fail to demonstrate some of these antibodies.

1. The saline agglutinin may be detected by the classical tube technique employing saline dilutions of serum with saline saline suspended red cells. The agglutinin also reacts in hydrophilic colloids such as bovine albumin and human plasma or serum and also gives a positive Coombs' developing test.[2] In addition, the agglutinin shows a positive reaction in both the capillary[1] and on the Diamond-Abelson slide tests.[3] This antibody which reacts in the presence of saline is found in the gamma globulin fraction of the antiserum. Since the agglutinin is inactivated by the various agents which are capable of denaturing gamma globulin, such as heat, lowered pH, etc., it is the most liable of the antibodies.
2. The agglutinoids or true blocking antibodies[15] not only fail to produce agglutination in saline but coat the antigen in such a manner that agglutination is inhibited when an antiserum with known saline agglutinins is added to this serum-cell mixture. The blocking antibody gives a negative saline

ORDER OF ANTIBODY ACTIVITY	Serological Tests						
	SALINE TUBE TEST	BLOCKING TEST	DEVELOPING TEST	ALBUMIN TEST	HEMOLYTIC TEST	CAPILLARY TEST	LOCATION IN SERUM GLOBULIN
AGGLUTININ	+	—	+	+	+	+	Gamma
AGGLUTINOID (TRUE BLOCKING ANTIBODY)	—	+	+	PROZONE OR +	+?	+?	Water Soluble
CRYPTAGGLUTINOID	—	—	+	+	+	+?	Beta and Water Soluble

Classification of Rh Antibodies

According to their serological and chemical behavior

Fig. 1

agglutination test, a positve blocking test, usually a prozone in the albumin test, and a positive Coombs' developing test. The capillary and slide test may or may not be positive depending on the other antibodies present. This agglutinoid is found in the water-soluble globulin fraction. It can be reversibly inactivated by some chemical reagents, but otherwise is quite stabile.

3. The cryptagglutinoid[7,8,9] neither agglutinates in the presence of saline nor does it inhibit agglutination in the blocking test, but it does give a positive Coombs' test and a positive Diamond-Denton[4] albumin test. In more than 50% of the mothers of erythroblastotic babies, this antibody is the only one detected during the pregnancy.[9] The cryptagglutinoids are found in both the beta and water-soluble fractions of serum.

Previously, the classical saline tube technique, which is used so widely in most serological procedures, was the only method used to detect the Rh antibodies. However, it soon became apparent that not all immunized individuals give a positive agglutination in titrations in which serial dilutions of serum plus saline suspended erythrocyte antigens are employed.

The percentage of antibody detections may be increased some-

what through the use of the true "blocking test." After the usual saline titration has been done and observed, a drop of serum known to contain saline agglutinins is added to each tube in the titration. The tubes are then incubated a second time at 37° C for 30 minutes. The failure to produce agglutination indicates the pressure of an inhibiting substance, the blocking antibody.[15]

The third order of antibody activity, the cryptagglutinoid, may be detected by the Coombs' test, Diamond-Denton albumin test, human serum, human plasma, or the albumin-plasma methods. Gum acacia, Le Page's glue and other hydrophilic colloids have been recommended in antibody detection procedures for the cryptagglutinoid, but these methods have been abandoned because of the possibility of non-specific clumping. The Coombs' test, Diamond-Denton bovine albumin titration, and the human plasma and albumin-plasma techniques give approximately the same range of sensitivity with the Coombs' test usually giving the higher titers. The albumin and plasma methods, however, are inadequate for one tube screening tests since many of the patients' sera contain a blocking antibody and might give a false negative result due to the blocking prozone.

The Coombs' test, although time consuming, has remained the most adequate test for one tube qualitative screening procedure and for the most precise of the quantitative titration procedures. The very nature of this test is such that it detects antibodies in general rather than one of specific activity. The Coombs' developing serum being anti-human globulin, reacts with any human globulin whether or not it is adsorbed on the test cells. To be sure that the developing serum reacts only with the antibodies directed towards the antigens employed, it is necessary to carefully wash the cells by centrifugation, at the end of the initial incubation period. The washing removes all serum proteins that are not specifically adsorbed on the red cells.

The Coombs' developing test has additional value as a diagnostic procedure for erythroblastosis fetalis. By this method it is possible to detect antibodies adsorbed on the erythrocytes of the affected infant. These antibodies damage the infants cells and thereby produce this disease. To perform this test, a drop of a two percent suspension of baby's red cells in saline are washed three times with saline by centrifugation. To these washed packed erythrocytes is added one drop of developing serum. This mixture is then incubated at 37° C for 30 minutes to one hour. At the end of this incubation period, the tube is centrifuged and examined for agglutination. Clumping is considered a positive test for adsorbed antibodies on the cells tested.

Recently, the capillary test has been recommended as an antibody screening technique. However, in view of the fact that the capillary test fails to detect all antibodies, the use of this method

to screen bloods for isoimmunization may be a dangerous procedure. This test is good when used in its proper place, that of detecting Rh antigens with a serum which has been standardized for that purpose. It should be emphasized that not all antibody producers will demonstrate a positive capillary test, particularly those individuals who produce only block and cryptagglutinoid antibodies. The speed of reaction (avidity) in immunized persons showing capillary test activity varies considerably between patients. Some sera may give visable reactions in a few seconds, while others may be as slow as 15-30 minutes. For these reasons, the use of the capillary technique for the detection of immunization in obstetrical and transfusion patients is an undesirable practice.

The slide technique has been proposed as a qualitative test for immunization against the Rh antigens. Pure saline agglutinins react poorly with this test. In addition, it is frequently difficult to obtain positive reactions with low titered cryptagglutinoid containing serum. If this test is used, its limitations should be carefully considered.

One of most recent of the antibody detection procedures employs trypsin-treated red cells as antigens. The techniques suggested are modifications of the test designed by Morton and Pickles.[13] Wheeler, Scholl and Luhby,[14] and Kellner, Haber and Hedel[10] have offered such modifications. The original Morton and Pickles method in which erythrocytes were treated with 1-400 trypsin for 20 minutes and resuspended as a 2% saline suspension seems to give the most sensitive titration.

Comparison titrations using albumin, the Coombs' test, and the trypsinated red cell techniques were done on 30 sera known to contain antibodies.[5] These methods (Figures 2 and 3) showed similar results with respect to the titration end-points. The trypsin-treated red cells were not consistently agglutinated in the low dilutions of sera known to contain agglutinoid (blocking antibodies), although this prozone was somewhat reduced as compared to the results observed in albumin titrations.

The Kellner, Haber and Hedel modification of the trypsin-treated red cell technique was offered as a fast cross match method and as a one tube screening test for the detection of Rh antibodies. This procedure consists of the addition of a 2% saline suspension of erythrocytes to two drops of the patient's serum. The suspension is then centrifuged immediately and observed for agglutination. After the preliminary observation, two drops of 2% trypsin (Difco) in saline is added to the serum-cell mixture. At the end of 5 minutes incubation at 37° C, the test is centrifuged a second time and observed for agglutination. This cross match procedure gives considerably more sensitive results than the conventional cross match using saline-

TEST	Dilutions of serum									
	1	2	4	8	16	32	64	128	256	512
Saline tube	4+	4+	4+	4+	4+	4+	+	-	-	-
Albumin	4+	4+	4+	4+	4+	4+	+	-	-	-
Blocking	-	-	-	-	-	-	-	-	-	-
Developing	4+	4+	4+	4+	+	-	-	-	-	-
Trypsin treated rbc. - 10 min.	4+	4+	4+	4+	4+	+	+	+	-	-
Capillary		1'50"	3'	4'	5'40"	17'	26'	45'	-	-
Slide	2'30"	3'	5'	6'	9'	-	-	-	-	-

Comparison of antibody titration methods

Serum-1 (essentially agglutinins)

TEST	Dilutions of Serum												
	1	2	4	8	16	32	64	128	256	512	1048	2096	4192
Saline tube	—	—	—	—	—	—	—	—	—	—	—	—	—
Albumin	—	—	—	—	+	2+	3+	3+	3+	2+	+	+	—
Blocking	4+	4+	4+	4+	4+	3+	+	—	—	—	—	—	—
Developing	4+	4+	4+	4+	4+	4+	4+	4+	3+	2+	+	+	—
Trypsin treated rbc.-10 min.	+	+/—	—	+	2+	2+	3+	2+	+	—	—	—	—
Capillary		1'15"	2'10"	3'	4'30"	—	—	—	—	—	—	—	—
Slide	30"	1'20"	1'35"	2'	5'15"	6'10"	7'	7'40"	8'20"	10'	11'	13'	—

Comparison of antibody titration methods

Serum-2 (blocking and cryptoagglutinoid)

Figs. II and III

cell suspension, plasma-cell suspension or serum-cell mixture.

However, in the examination of 55 serum samples showing various known antibody patterns[5] by the Kellner, Haber and Hedel technique, negative results were observed in eight sera from five patients known to contain blocking antibodies having titers ranging from 1/64 to 1/256, and cryptagglutinoids of 1/1000 to 1/512,000. Six of the sera examined contained a low titered anti-M antibody which gave a positive reaction in the saline-cell-serum stage of this cross match, but gave negative results after the addition of trypsin. The only false positive reactions found in a series of 200 routine blood samples were in two patients known to have cold agglutinins.

After the above results were obtained a study was made on the action of trypsin in the cross match test. Since our previous experiments on the physico-chemical properties of antibodies demonstrated that the time of action of reagents on globulins is an important factor, it was felt that possibly some of the false negative results might be due to the shortness of time during which the trypsin was allowed to react. Consequently, several variations of the trypsin cross match test were studied. It was found that an increase in the incubation time after the trypsin was added yielded more satisfactory results.

The best results were obtained by the following method. After the addition of the suspension of cells to the patient's serum in the first step of the test, the mixture was allowed to stand 5 minutes at 37° and was then centrifuged at 1000 r.p.m. for one minute. After the preliminary readings, two drops of 2% trypsin was added to the tube and incubated for 30 minutes at 37°.

With one exception, the eight sera containing a high concentration of agglutinoids, gave 2 to 4 plus positive agglutination results. Only one serum reacted poorly giving a 1 plus agglutination. The six samples of anti-M isoimmunization sera remained negative by this method. From this data, it is apparent that the additional 30 minute incubation is well worth the time spent since it provides greater sensitivity and safety in the cross match. Although time is frequently limited in the clinical laboratory, the importance of the cross match is so great that the extra time allotted is indicated. Accuracy is more important than speed in a transfusion service.

Although the trypsin treated red cell technique gives good results in the detection of antibodies, it has its limitations. It should be emphasized that the method is not reliable in testing for adsorbed antibodies on the red cells of erythroblastotic infants. This procedure can not be substituted for the Coombs' developing test for the diagnosis of erythroblastosis.

DISCUSSION

All the antibody detection procedures discussed have their place when properly used and properly interpreted. Fortunately, some sera will give positive results by almost any test as shown in Figure 2. However, each test has its limitations as well as its uses. Figure 3 illustrates a serum in which a battery of tests was needed to show the true antibody pattern.

Certain precautions should be observed in the performance of antibody determinations regardless of the testing procedures chosen.

1. Glassware must be chemically clean. Small amounts of acid or soap may denature or interfere with the antibody globulin reactivity.

2. Reagents must be good quality and free of bacterial contamination. Growth of bacteria in serum may produce either false positives from bacteriogenic agglutinins or false negative reactions due to bacterial interference.

3. The erythrocytes used as the test antigen must be relatively fresh and free of bacterial contamination. The maximum antigenicity of a cell is about four days. The genotype of these cells should be determined since the specificity of the test depends on the antigens possessed by the cells employed.

4. Reference sera to be used as controls should be kept frozen when not in use and should be tested occasionally with cells of known antigenic components. In this way, the strength and specificity of the control serum may be determined.

5. Care must be taken in centrifugation so that the tubes are not spun too long or too fast. (1000 r.p.m. for one minute is adequate.) Prolonged centrifugation may result in centrifuge clumping which is sometimes hard to distinguish from true agglutination.

6. In reading tests, it must be remembered that the very nature of the antigen-antibody agglutinate is such that it is easily dispersed by rough handling. For this reason, it is possible to shake a 3 or 4 plus reaction in such a manner so that the test appears negative.

By observing a few simple precautions and understanding the limitations of each method, the technologist should have no more difficulty in performing these tests for blood antigens and antibodies than in the performance of any other routine clinical laboratory procedure.

REFERENCES

1. Chown, B.: A rapid, simple and economical method for Rh agglutination. Am. J. Clin. Path. 14: 114-115, 1944.
2. Coombs, R. R. A., Mourant, A. E., and Race, R. R.: Detection of incomplete Rh agglutinins. A new test. Brit. Exper. Path. 26: 255-266, 1945, Lancet 2: 15-16, 1945.
3. Diamond, L. K., and Abelson, N. M.: The demonstration of anti Rh agglutinins. An accurate and rapid slide test. J. Lab. Clin. Med. 30: 204-212, 1945.
4. Diamond, L. K., and Denton, R. L.: Rh agglutination in various media with particular reference to the value of albumin. J. Lab. Clin. Med. 30: 821-830, 1945.
5. Guy, R., Patel, T. D., Hill, J. M., and Haberman, S.: Unpublished data.
6. Haberman, S., Hill, J. M., and Reid, A. E.: Some physical and chemical aspects of Rh antibodies.: In press.
7. Hill, J. M., Haberman, S., and Guy, R.: Further evidence for third order antibodies. Fractionation of agglutinins, blocking antibodies, and cryptagglutinoids by physico-chemical methods. Am. J. Clin. Path. 19: 134-140, 1949.
8. Hill, J. M., Haberman, S., and Jones, F.: Hemolytic Rh immune globulins: Evidence for a possible third order of antibodies incapable of agglutination or blocking.: Blood, J. Hem. 3 Supp. 80-110, 1948.
9. Hill, J. M., Reid, A. F., and Haberman, S.: The Rh antibody problem. Differentiation of agglutinins, agglutinoids, and cryptagglutinoids. Tex. State Med. J. 45: 477-481, 1949.
10. Kellner, A., Haber, J. M., and Hedel, E. F.: The detection of Rh blocking antibodies in routine cross matching by means of trypsinated red cells. Proceedings of the American Association of Blood Banks, Second Annual Meeting. 49-55, 1949.
11. Landsteiner, K., and Wiener, A. S.: Agglutinable factor in human blood recognized by immune sera for rhesus blood. Proc. Soc. Exper. Biol. and Med. 43: 223, 1940.
12. Levine, P., and Stetson, R. E.: Unusual case of intragroup agglutination. J. A. M. A. 113: 126-127, 1939.
13. Morton, J. A., and Pickles, M. M.: Use of trypsin in the detection of incomplete anti Rh antibodies. Nature 159: 779-780, 1947.
14. Wheeler, W. E., Scholl, M. L. L., and Luhby, A. L.: The use of trypsin treated red cells for the detection of anti Rh antibody. Health Center Jour. O. S. U. 2: 86, 1949.
15. Wiener, A. S.: A new test (blocking test) for Rh sensitization. Proc. Soc. Exper. Biol. and Med. 56: 173-176, 1944.

A CASE OF FULMINATING MULTIPLE MYELOMA WITH MASSIVE HEMORRHAGE

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This study and review was prompted by a fulminating case of multiple myeloma with massive bleeding, renal in origin, in a young man, 37 years of age. The duration of the illness was ten weeks from the onset of symptoms to death.

B. S., a 37-year-old, married, college student entered Memorial Hospital October 31, 1949, complaining of severe pain in the right chest of six weeks duration and gross hematuria of three days duration. The pain was almost continuous in nature and one week prior to admission had radiated down into the sacro-iliac region. The onset of the hematuria was abrupt with grossly bloody urine at every micturition. Physical examination revealed essentially normal conditions except for severe bilateral conjunctivitis and pain in the costovertebral regions on deep palpation. No external cause for the chest pain was noted.

On admission to the hospital, routine blood count revealed moderate anemia and severe granulocytopenia, while urinalysis showed albuminuria and gross hematuria. The hemogram was as follows:

RBC—3,380,000	Seg.—35%
WBC—3,550	Lymph.—63%
Hb—10 gms.—65%	Eosin.—2%

The following day, a cystoscopic examination was done, and the specimen collected showed a large volume of red cells; and the urogram was suggestive of polycystic kidneys, although not typical. The patient continued to bleed profusely from his kidneys and intermittently from the nose. Laboratory examination showed a marked hyperproteinemia with the A-G Ratio reversed, a marked nitrogen retention. When the serum was centrifuged for the chemistries, a marked autohemagglutination was noted. The prothrombin and clotting time were prolonged. The Bence Jones was found to be positive. The formol-gel test was also positive. Both the Bence Jones and formol-gel are significant of multiple myeloma; but because of the profuse renal bleeding, which is unusual in multiple myeloma, the final diagnosis was made after a bone marrow study was done. The bone marrow study showed myeloma cells and thus definitely established the diagnosis.

The patient became progressively worse throughout his hospital stay; and supportive therapy including blood transfusions, Protamine Sulfate for a possible hyperheparinemia and paren-

teral fluids were given with no appreciable result. After three weeks' hospitalization, the patient showed myeloma cells in the peripheral blood, and a few nucleated red blood cells. Several small areas of decreased density of the skull and ribs were revealed by X-Ray examinations, were considered suggestive of multiple myeloma by the radiologist. The right fifth rib was now found to be irregular in outline. The laboratory procedures done were varied, and every phase of the disease was checked as thoroughly and as often as possible.

The findings were those typical of multiple myeloma; the hyperproteinemia; nitrogen retention, Bence Jones and formolgel were positive. The nitrogen retention was decreased somewhat but still persisted after a great deal of treatment. Blood was found in the urine constantly, varying in amount. Although it was almost impossible to entirely free the urine of blood used for the Bence Jones test, making the reading difficult, the test was definitely positive on several occasions. The blood consistently showed autohemagglutination. It was found extremely hard to centrifuge even though the patient's cell count was low in proportion to the serum. Invariably, centrifuging several times at high speed was necessary to separate the clot and the serum. The patient persistently showed an hemolysis of the serum and sometimes an elevated icterus index. There were changes in the liver metabolism, prolonged prothrombin time, but the platelets were normal. The A-G Ratio was reversed, the amount of globulin greatly increased in comparison to the albumin.

The blood smear on admission showed no abnormality except for the presence of lymphocytes and leukopenia, but the lymphocytes were normal in appearance. However, as the disease progressed, the lymphocytes remained; but in addition, myeloma and Turk cells were noted which correlated with the bone marrow findings; these findings were conclusive evidence for the final diagnosis of multiple myeloma, the bone marrow revealing a large percentage of myeloma cells, characteristic of the disease. The sternal aspiration was difficult. The slides which were obtained were consequently very poor as only a small amount of bone marrow could be aspirated. The patient experienced a great deal of pain, and the site of puncture remained open and bled until his death, which occurred 11/30/49, one month after admission to the hospital and ten weeks after onset of the disease.

The autopsy findings were consistent with the diagnosis. The lungs showed a few old adhesions; and when removed, the thoracic cage revealed a nodular growth involving the 7th rib at the axillary line. There were a few small nodular areas on the ribs in the left side of the chest. The lesion on the right side was removed; and when sectioned, there was a large lesion of

DATE	Prothrombin	NPN	Creatinine	Urea Nitrogen	Uric Acid	Total Protein	Albumin	Globulin	A-G Ratio	Icterus Index	Bromo Sulphalein	Chloride	Formol-Gel	Sugar
11- 2-49		55mg %	1.75mg %											130mg %
11- 8-49	Pt-18 sec Cont-16 sec	160mg %	8.3mg %			12.9gm %	2.3gm %	10.6gm %	0.2 tol	12 Units		660mg %		
11- 9-49													Positive	
11-12-49		170mg %	8.3mg %		10.3mg %	9.15gm %	2.3gm %	6.85gm %	0.3 to 1					
11-14-49	Pt-32 sec Cont-16 sec	96mg %	6mg %							5 Units				
11-15-49						10.45gm %	5.1gm %	5.35gm %	0.9 to 1		5 min-10% Retention 30 min-0% Retention			
11-17-49	Pt-45 sec Cont-16 sec	73mg %	3.6mg %	54mg %	6mg %	10.4gm %	2.2gm %	8.25gm %	0.2 to 1				Positive	127mg %

DATE	Red Blood Count	White Blood Count	Hemeglobin	Seg	Stab	Juv	Eosin	Baso	Lymph	Prolymph	Myeloma	Türck	B1&C1	Platelets	Transfusions
10-31-49	3,380,000	3,550	10gm—65%	30	5		2		63						
11- 8-49	2,470,000	6,150	8.5gm—55%	25	6		Nucleated RBC-2		69				B1-30 sec. C1-10 min.		
11- 9-49	710,000		Unable to read											200,000 per Cu mm	500 cc Whole Blood
11-12-49	2,700,000	7,050	8gm-50%	25	9		2		64						500 cc Whole Blood
11-14-49	3,200,000	8,400	7gm—45%	39	10				51						500 cc Whole Blood
11 16-49	2,550,000	9,700	9.5gm—63%	29	9	6	1		51	5					500 cc Whole Blood
11-21-49	1,745,000	7,400	Unable to read	14	12		1	1	56		15	1			

		Stab	Lymphocyte	Lymphoblast	Myeloma	Nucleated RBC
11-17-49	Bone Marrow	4	35	1	52	6

DATE	Color-Character	Reaction	Specific Gravity	Albumin	Sugar	Acetone	RBC	WBC	Epithelial Cells	Bence Jones
11- 1-49	Dark Red Cloudy	5.5	1.010	4+	Neg.	Neg.	Loaded			
11- 6-49	Black Cloudy	5.5	1.016	4+	Neg.	Neg.	Loaded			
11-14-49							Packed			Positive
11-21-49	Amber Cloudy	5.0	1.012	4+	Neg.	Neg.	20—30 H P F	Few		
11-29-49										Positive

the medullary cavity of the rib. The tissue appeared grayish and gelatinous in character; and there was a loss of normal cortex of the bone. The sternum was sectioned lengthwise, and the sternal marrow appeared pinkish-red in color without any definite tumor formation. The lungs on cut section showed hypostatic congestion and had a very pale appearance, the pallor due to the marked anemia that the patient suffered. The spleen was markedly enlarged, measuring 10 x 8 x 2 inches. On cut section, there was the typical appearance of splenic tumor, being soft in character and bulging on cut section. The kidneys were similar on both sides, both slightly enlarged. The capsules stripped readily. The kidney pelves externally showed a reddish-brown color; when opened, the entire pelvis of both kidneys presented the appearance of a superficial layer of clotted blood, overlying and adherent to the normal epithelium of the kidney pelvis, and could not be scraped off. The kidney substances showed normal differentiation between cortex and medulla. Although the cortex was slightly narrow, no areas of petechiae nor hemorrhage were seen and no gross changes noted.

Microscopic sections of lung showed red cells; and in one area, there was a definite localized collection of myeloma cells. Sections of kidney showed a normal appearing glomeruli; many of the tubules showed amorphous material plugging the lumen, and there was scattered throughout a localized collection of myeloma cells. Beneath the pelvis epithelium, there was evidence of old and recent hemorrhage with considerable laking of cells with an infiltration of phagocytic cells. Microscopic sections of spleen were essentially normal except for engorgement of the splenic sinusoids with red blood cells. Sections of bone marrow showed definite tumor formation. This tumor formation consisted of packed myeloma cells extending out into the fibers of bony structures, and on the outside of the rib there was a layer of this myeloma cell tissue.

In reviewing studies by Boyd and Heck of the Mayo Foundation; by C. H. Geschichter, of Johns Hopkins; by Drs. Ralph Ghormly, Pollock, Byron Hall and Lawrence Biezer of Rochester, Minn., it is found that the disease has an insiduous onset, with the fulminating type occuring rarely. The early symptoms are pain, swelling and pathological fracture. The age group has been found to be from 40 to 60 years. Our case was 37 years, exceptionally young for this disease. The renal complications are nitrogen retention, albuminuria and Bence Jones proteinemia. The bone marrow is stuffed with myeloma cells. The peripheral blood shows a leukopenia and a lymphocytosis at onset and later shows myeloma cells and immature red cells. Anemia has been noted resulting from invasion of the bone marrow. X-ray shows "punched-out" defects where the bone destruction occurs. This is especially seen in the skull and ribs. Abnormal bleeding is largely from the nose and gums. The high protein noted is due to the excess formation of globulin.

In comparing the case presented with previous studies, the typical findings reported in the above reviewed literature were found, but in addition, the striking features in this case were the massive hemorrhage from the kidney, nose and sternal puncture site; the short duration of the illness in a comparatively young man, and the fulminating type of the disease.

BIBLIOGRAPHY

Christopher, Frederick: Textbook of Surgery—Multiple Myeloma. 5th Edition. Saunders pp. 187.

Shands, A. R.: Multiple Myeloma, Hand Book of Orthopedic Surgery. 3rd Edition—Mosby—pp. 273.

Ghormly, Ralph; Pollock, Geo.; Hall, Bryon; Beizer, Lawrence: Surgery, Gynecology and Obstretrics—Vol. 74—Jan.-June, 1942.

Bayard, Edwin; Heck, Frank J.: Review of 83 Proved Cases. Vol. 133. No. 3.—J A M A.

Geschichter, C. A., and Copeland, M. M.: Multiple Myeloma. Arch. Surg. 16:807; April, 1928.

COLORIMETRY: A BRIEF SURVEY OF PRINCIPLES

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Lord Kelvin once said, "I often say that when you can measure what you are speaking about and express it in numbers, you know something about it; but when you cannot express it in numbers, your knowledge is of a meager and unsatisfactory kind: it may be the beginning of knowledge, but you have scarcely, in your thoughts, advanced to the stage of science, whatever the matter may be." We have today a science of color. Color is something which can be measured and expressed in numbers. The science of color measurement is called colorimetry.

Instrumental procedures in chemical analysis may depend on compartive observation of the intensity of color produced in a system absorbing visible light. Or it may depend on an absolute measurement of the amount of light absorbed by the system. The procedures in the first of these include the long-established so-called colorimetry of chemists. This is a misnamed but well understood term which has come to mean chemical analysis depending on the comparison of the color of the substance under investigation with the color of a standard. Recent emphasis, however, has been on the development of rapid methods of measurement of the light absorbed by the analytical system, that is, of photometric methods. This paper is a brief outline of the fundamentals of photometry, a science that applies the laws of light absorption to analytical chemical measurements.

Let us review briefly the laws of the absorption of light. Lambert's law states that when a parallel beam of monochromatic light passes through a homogeneous absorbing medium, the light intensity diminishes exponentially with the thickness of the absorbing medium. Thus each layer of equal thickness of absorbing matter absorbs an equal fraction of the light that strikes it. The size of this fraction is a function of the inherent capacity of the absorbing medium to absorb the monochromatic light in question. If layers of the thickness of a single molecule are considered, it may be said that each molecule absorbs an equal fraction of light.

It is convenient to distinguish between intensities of the original or incident light and the transmitted beam by the respective symbols I_o and I_x. Then,

$$I_x = I_o e^{-kd}$$

where I_x is the intensity to which the original intensity, I_o, of a parallel monochromatic beam is reduced after passage through a thickness, d, of absorbing medium; and where k is the absorption coefficient, a constant for a given wave length, but, in general, a function of wave length and temperature; and where e is the base of the natural logarithms.

The above equation may be expressed logarithmically, to the base 10:

$$\log \frac{I_x}{I_o} = -K\,d$$

or:

$$\log \frac{I_o}{I_x} = Kd$$

where K is k/2.303 and is usually termed the extinction coefficient.

The ratio I_x/I_o is the fraction of light transmitted by thickness, d, of the medium; and the log of its reciprocal, log I_o/I_x, is the optical density of the medium.

In the simple case in which the molecules of an absorbing substance act independently of each other, and in which the absorption of the solvent does not change with concentration, each molecule of the absorbing substance transmits the same fraction of incident light as each other molecule of that substance. Thus the light transmitted decreases exponentially with increasing concentration of the absorbing substance.

We now have Beer's law for monochromatic light:

$$I_x = I_o e^{-k_m cd}$$

where c is the concentration in moles per liter, d is the thickness in centimeters and k_m is the molar absorption coefficient, a characteristic of the absorbing substance, but depends on solvent, wave length, and temperature.

Experimental results are usually reported in terms of the molar extinction coefficient, ε, defined by the relation:

$$\log I_o/I_x = \varepsilon cd$$

or

$$\varepsilon = D/cd$$

where D is the optical density, c is the concentration expressed in moles per liter, and d is the thickness expressed in centimeters.

In the same way, the calibration or specific extinction coefficient is defined by the relation

$$K = D/cd$$

where c is the concentration in grams per milliliter.

Since

$$I_x/I_o = T$$

where T is transmitted light, and if d = I, then,

$$\varepsilon = -\log T$$

This becomes:

$$\varepsilon = \log \frac{100}{T} = \log 100 - \log T$$

when the reading of the transmission on a photometer scale is from 0 to 100%.

Then the value of a solution of unknown concentration may be calculated from the formula:

$$C_u = \frac{C_s \times \varepsilon_u}{\varepsilon_s}$$

where C_u is concentration of unknown solution, C_s is concentration of a known solution.

The usual working formula for a system in which the value of K has been determined by a calibration curve is:

$$C = \frac{\log 100 - \log T}{K}$$

or:

$$C = \frac{2 - \log G}{K}$$

where C is concentration of the unknown colored solution, G is galvanometer reading and K is the calibration constant.

To test the applicability of Beer's law, the densities of solutions of the same thickness are plotted against their concentrations. A straight line passing through the origin shows that Beer's law holds true, the slope being equal to the product εd. It is often obeyed; exceptions indicate molecular interactions, such as those causing association, dissociation, complex formation, or change in solvation. It frequently happens that with light filtered from a white source, linearity is obtained at lower concentrations, with deviations at higher concentrations.

If two colored substances, each obeying Beer's law, are present in the same solution, their concentrations can be determined by measuring the absorption at two wave lengths, for which the respective molar extinction coefficients are known. If the absorption bands are far apart, measurement with the use of sharp filters at the two maxima will permit the calculation of the concentrations from the regular colorimetric formula. Even if the bands overlap, measurements at two wave lengths at which the separate molar coefficients are markedly different in magnitude will allow the concentrations to be determined. If D′ and D″ are the measured densities of substances 1 and 2 at wave lengths λ' and λ'', c_1 and C_2, the concentrations of the absorbing substances, ε_1' and ε_1'' the molar extinction coefficients of 1 at λ' and λ'' and ε_2' and ε_2'' similarly the molar extinction coefficients of 2, and d the thickness of the absorbing layer, then

$$D' = (\varepsilon_1' c_1 + \varepsilon_2' c_2)d$$

$$D'' = (\varepsilon_1'' c_1 + \varepsilon_2'' c_2)d$$

from which

$$C_1 = \frac{\varepsilon_2'' \cdot D'/d - \varepsilon_2' \cdot D''/d}{\varepsilon_1' C_2'' - \varepsilon_2' \varepsilon_1''}$$

and

$$C = \frac{\varepsilon_1'' \cdot D'/d - \varepsilon_1' \cdot D''}{\varepsilon_1'' \varepsilon_2' - \varepsilon_2'' \varepsilon_1'}$$

Sensitivity of photometric methods is increased by filters transmitting only the light absorbed by the substance to be analyzed. The position and intensity of the absorption band may be determined by the rather elaborate procedure of plotting a spectral curve, or by the simplier spectrometric examination using such an instrument as a hand spectroscope. Knowledge of the position of maximum absorption permits selection of the best filter. The filter is chosen which has maximum transmission at or near the absorption maximum of the substance to be analyzed. The intensity of the band determines the sensitivity of the method.

A factor in the over-all accuracy of a colorimetric method depends on the precision of the photometric measurement. It can be shown that a given error in the transmission causes less error in the concentration at a density of 0.434 or 36.8% transmission, but the minimum in error as a function of transmission is flat, so that from 20% to 60% the accuracy is not materially different. On either side of these limits, however, the error rises rapidly and for most precision the concentration should be arranged so that transmission never falls outside the limits of 10% and 90%.

An otherwise suitable colorimetric system need not be discarded if it shows deviations from Beer's law. However, one should make sure that nonlinearity is really intrinsic and not merely due to a poor choice of filter or some other correctable factor.

When Beer's law is not obeyed, K is not a constant, and a calibration curve must be used for calculations. In this curve the per cent transmission is plotted against concentration from data obtained from analysis of standard solutions. Concentration is then read directly off the curve for any value of transmission. A recalibration following any change in the system, however, will require the determination of several points. Only with complete standardization in respect to light source, filter, and receiver would it be thinkable to use a curve obtained with other apparatus as the basis of a determination. And in any circumstance proof of standardization would demand verification of at least a few points on the curve.

THE USE OF FILTER PAPER AS AN AID IN THE ISOLATION OF S. TYPHOSUM

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A paper was published by Bedson and Bland (1929)[1] describing the electronegative property of filter paper, and how this property could be utilized for determining the charge carried by virus particles. These workers used strips of blotting paper 18 cm. long, by 0.5 cm. wide, marked off in one cm. divisions. The strips were held in a clamp, attached to a burette stand. The lower 0.5 cm. was allowed to dip into a wide mouthed dish, containing the virus suitably diluted with a requisite buffer. The fluid was allowed to rise to the 10 cm. mark, the strip then withdrawn and cut into 1 cm. sections, each of which was dropped into a numbered sterile tube. These samples were then examined for the presence of the virus by animal inoculation.

This work was noted by Colquhoun (1930),[2] who applied the same principles in the investigation of bacteria of the coli-typhoid group. The authors of this paper refer to the well known fact that by cataphoresis, bacteria are shown to cary a negative sign or charge, and that differences in the degree of charge can be determined by observing their rate of migration across an electric field. This led to the belief that similar differences might be shown by testing the passage of bacteria into filter paper. This was found to be so, using the commoner members of the coli-typhoid group. E. coli when mixed with paratyphoid-salmonella group of organisms in dilutions of 1-2000 could be recovered from the filter paper fairly readily.

In both of the above publications reference is made to the fact that staining solutions with opposite charges, such as methylene blue and eosin, behave similarly. If solutions of these stains be dropped onto filter paper, the methylene blue remains localized and will not pass into the paper, whereas the eosin diffuses readily. Bedson and Bland, employing their filter paper technique with the composite Leishman stain, demonstrated the negative charge carried by the filter paper, by the fact that the eosin travels up the strip with the water a considerable distance, whereas the methylene blue remains constantly at little more than one cm. above the level of the stain contained in the dish. The particles of methylene blue are positively charged leading to a firm adsorption of the stain on the paper and thus prevented from passing along with the water. The eosin, carrying the negative charge, has no such barrier.

The work about to be described contains nothing of an original nature, and apart from a few additional observations and modifications, the experiments largely confirm the work of Colquhoun. The many tests made have been confined to S. typhosum, strain TY2. Other members of the paratyphoid-salmonella groups have not been investigated so far.

A search of the literature has proved unsuccessful in tracing other references to this subject. The method of employing filter paper with its negative charge, allowing only particles with a similar charge to pass into it, whereas the opposite charged particles are held in a state of absorption at the bottom of the paper, would suggest a simple method of isolating specific organisms in many fields, in which similar problems may be encountered.

SELECTION OF FILTER PAPER

In the previous publications, the choice of the paper used for conducting the tests were confined to blotting paper and Whatman filter paper, but no mention of the grade or porosity of such paper was mentioned. Accordingly several grades of blotting paper, were tested, and in addition a wide range of the various porosities of the Whatman paper to evaluate the relative merits of each paper. In our hands the most uniform results were obtained from the Whatman No. 1 paper, and this was used throughout in the tests to be described.

One curious observation was made with the tests employing ordinary office blotting paper. The three types of such paper readily available at this centre were in the colours, white, red and green. Filter paper strips of the red, and green, dipped into mixtures of E. coli, and S. typhi, gave no growth of these organisms on subsequent sub-culture. Presumably the dye content of these papers was bacteriostatic and growth of the organisms was inhibited.

THE METHOD OF SETTING UP THE TEST

Whatmans No. 1 filter paper was prepared by cutting into lengths of 30 cm., by 1 cm. in width. These strips were marked off into ½ cm. sections and such sections were removed and sampled by the appropriate methods (see later). The strips prior to use were sterilized in the hot air oven.

For use, a sterilized strip is held in a bulldog clip, which is held in a clamp fixed to the top of a burette stand. At the base of the stand, a small petri dish is fixed into position by the aid of plasticine. The dish will contain the prepared suspensions of organisms to be tested. The strip is now adjusted so that the

lower ½ cm. only will dip into the bacterial suspension, contained in the vessel at the base of the stand. The dish is left in this position for exactly 40 minutes (see later). At the expiration of this period a column of water will be found to have risen to a height of approximately 10½-11 cm. The height of the column of "wetting" is recorded. Samples of the wetted strip are now taken, using for this purpose sterile scissors and forceps. Commencing at the top of the wetted strip, the first ½ cm. section is removed, by cutting across the penciled mark previously mentioned. This is repeated down the entire wet strip. Each ½ cm. section so removed, is placed into a sterile petri dish, which is numbered in grease pencil appropriately.

The sections are now transferred from the petri dishes, to test tubes (numbered to correspond with the dishes) containing 5 ml. of peptone water. The sections are transferred to the peptone water, by the aid of a platinum loop, which is sterilized in the bunsen flame between each transfer.

The peptone water tubes, containing the ½ cm. sections, are incubated for 18 hours at 37° C. Sub-cultures are then made from the incubated samples onto McConkeys media, contained in petri plates.

PREPARATION OF THE TEST SUSPENSIONS

It was decided in the first instance to study the behavior of filter papers, of mixtures, in equal parts of E. coli and S. typhi suspended in isotonic saline. An arbitrary concentration of the organisms considered suitable were suspensions, diluted to opacity 3 (Brown's Opacity tubes).

Accordingly cultures of E coli and S. typhi were inoculated onto agar slopes and incubated at 37° C overnight. The overnight growth was removed from the slopes, by adding 2 ml. of isotonic saline and gently rubbing with a sterile loop. The dense suspension from both tubes was transferred to fresh tubes, from which further dilutions were prepared to match opacity 3.

For the test, equal parts of these suspensions were taken and thoroughly mixed by stirring with a sterile glass rod. Usually 2 ml. of each suspension was found to be adequate to give a satisfactory level, employing as a container a petri dish 6 cm. in diameter. The filter strip is then suspended in this mixture in the manner previously described.

USE OF A CLOSED CONTAINER

At the commencement of this series of experiments, the filter strips were immersed in the bacterial suspension and left for the

specified time on the laboratory bench, open to the atmosphere. This led to discrepancies in the height of wetting, giving a day to day variation. This was attributable to atmospheric changes affecting the evaporation of the fluid from the filter strip which was so exposed. To overcome this disadvantage the filter strip and suspension etc. was enclosed in a bell jar (see Fig. 1), the lid of which was sealed in position with "Apieson" sealing compound. By thus enclosing the apparatus in this manner, evaporation reaches an equilibrium, and controlled conditions of "wetting" are obtained.

Fig. 1

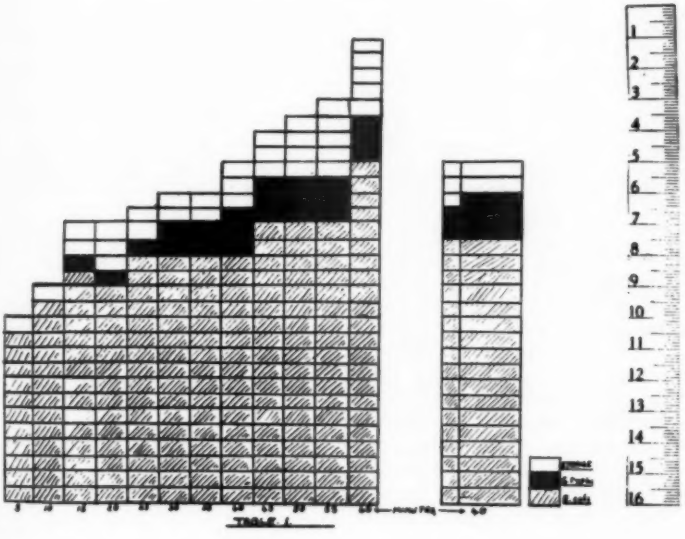
Filter Strip and suspension enclosed in "bell-jar." The fluid rises up the strip by capillary attraction. By enclosing the apparatus in the jar, evaporation reaches an equilibrium and controlled conditions are maintained.

STUDY OF THE EFFECT OF TIME FOR IMMERSION OF FILTER STRIP

The previous workers, took their samples at predetermined levels, i.e., Bedson and Bland removed sections when the column of wetting was 10 cm., whilst Colquhoun sampled at a wetting height of 2¼ inches. Both workers attained excellent results by their own method, but it seemed desirable to conduct a series of tests in an endeavour to establish whether there was any optimum time limit, for maximum recovery of specific organisms. Standard strips were therefore suspended for five minutes and the height of wetting recorded, and samples removed in the described manner. This was repeated at five minute intervals, so that samples were taken after immersion at 5, 10, 15, 20 etc. minutes, and so on to a period of one hour. At a later period 1½ and 2 hour immersions were tried, but this showed no advantage. The peak for optimum recovery, combined with the least waste of time was found to occur between 40-60 minutes immersion (see Table I). Times extended outside of these limits gave no increase in efficiency, the extended times however did prove that

a safety margin for sampling existed. If for any reason the operator was unable to remove samples at 40 mins., if they were recovered after this time pure cultures would be just as easily found; they would however be found higher up the strip, which would mean a consequent increase in the number of samples taken. It would appear therefore that if the sampling time is prolonged, the organisms of E. typhi occupying about a 1 cm. zone near the top of the strip are merely "pushed" higher all the time, by the coliform organisms, which still lag behind, but never apparently able to pass through the barrier formed by the typhoid organisms. This was supported by the long periods of immersion referred to above (1-2 hrs. etc.).

At this stage further tests were incorporated to decide the relative merits or otherwise of different widths of the strip, using such strips cut at 1½ cm. and ½ cm. wide showed no advantage (See Table I, the results obtained were identical to those obtained with strips 1 cm. wide.



THE STANDARD TEST

From the observation of the previous experiment, a filter strip was immersed for 40 minutes, into a suspension diluted to opacity 3 of equal parts of E. coli and S. typhi. This was repeated many times. The results obtained (see Table 2) were so regularly constant, that whenever the technique was varied, or the test was modified in any way, the "Standard Test" was always incorporated and it served as an excellent control. As will be seen from the Table the first two ½ cm. samples taken from the top of the wet strip always proved to be sterile. The following two to four ½ cm. sections will show a pure culture of S. typhi, no single colony of E. coli is apparent. The remaining ½ cm. sections from the remaining length of the strip will show a predominant growth of E. coli. Occasional colonies of S. typhi can be picked out, but such colonies are few in number. For this reason they are not recorded in the tables given throughout this paper.

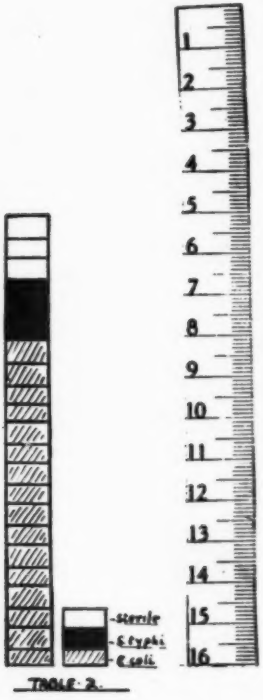


TABLE 2.

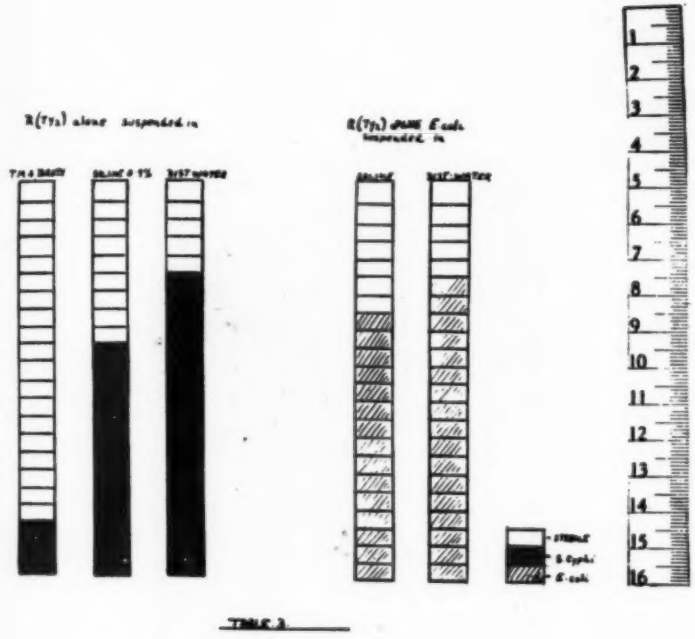
It will be noted therefore, that if a complete segregation of the two organisms be required, the filter paper technique would be the method of choice for this purpose.

ISOLATION OF ROUGH VARIANTS OF S. TYPHI

The previous experiments were conducted using the smooth strain of S. typhi (TY2). Colquhoun stated that organisms in the rough phase failed to pass into the filter strip.

He considered this due to the fact that rough variants are hyperagglutinable in isotonic salt solution. Passage into the filter paper was thought possible only if the concentration of the electrolyte was reduced, so as to overcome spontaneous agglutination. This view could not be supported by the present writers, because it was found that if a rough variant alone be suspended in isotonic salt solution and tested in the prescribed manner, the variant would pass into the strip for a distance of

8½ cm. (see Table 3). If however the rough strain be mixed in equal quantities with E. coli, then the rough strain will not pass into the paper, only the E. coli will be found, and recovered by the standard method of sampling. This result was obtained many times and it was quite conclusive that irrespec-

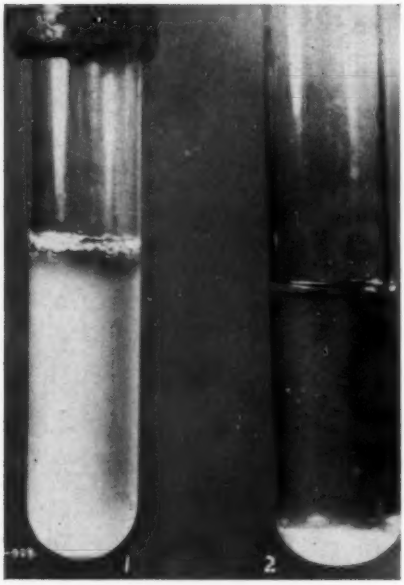


tive of the menstruum used as a diluent, (even the relatively electrolyte free, distilled water, if R(TY2) and E. coli be mixed together passage of the variant is always "blocked." It would appear from these observations that the so called electric charge is nullified in this particular test, and the answer is probably a mechanical one due to the mixture of the two organisms.

The method for obtaining the rough variant, and the subsequent tests are fully described as follows:

From an 18 hours growth of the Smooth strain of TY2, subcultures were made into 10 ml. of tryptic meat broth, containing

0.25 ml. of S. typhi immune sera. Daily sub-cultures were made into similar tubes of broth for twenty days. After this period the broth culture had changed from the dense cloudy suspension, to one showing a clear supernatent fluid, containing course aggregates, indicating a complete reversion to the rough colonial stage. From plate cultures, a typical rough variant was selected conforming strongly to type, (see Fig. 2) and which was markedly hyperagglutinable in isotonic salt solution. Using this variant, the standard test was carried out in the following man-



Obtaining Rough Variant TY2

Tube 1 shows homogeneous suspension of smooth type S. typhi. Tube 2 shows the same smooth strain after 20 serial sub-cultures in broth containing anti-O serum. Note the coarse floccules and deposit at the bottom of the tube, typical of the change from S. to R.

Fig. 2 (a)

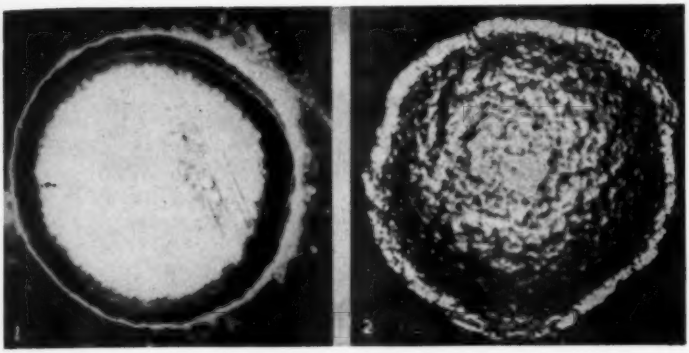


Fig. 2(b)
Plate (1) shows a smooth colony, sub-cultured from tube (1) above.
Plate (2) shows typical rough colony, sub-cultured from tube (2) above, after serial passage in broth containing anti-O sera.

ner. Suspensions were prepared and diluted to opacity 3 of Brown's tubes in the following fluids:

(1) 0.9% saline
(2) Distilled water
(3) An undiluted broth culture } R (TY2) alone

(4) 0.9% saline
(5) Distilled water } Containing in suspension equal parts of S. typhi and E. coli.

As will be seen from Table 3, the rough stain passes freely into the filter paper when suspended **alone** in 0.9% saline and slightly higher when suspended alone in distilled water, and to a lesser extent (but nevertheless still passing) with broth cultures undiluted. When the rough strain of TY2 is mixed with E. coli, however, the rough variant cannot be shown at any level on the strip, indicating that its passage is blocked, presumably by the presence of the E. coli.

ISOLATION OF R(TY2) BY FILTER PAPER, IN SUSPENSIONS CONTAINING A SURFACE ACTIVE AGENT

From the previous experiment in which it was shown that mixtures of E. coli and S. typhi when mixed prevented the passage of the R. strain. An attempt to overcome this barrier between the two organisms, was made by suspending the organisms in a solution containing a surface active agent.

For the purpose sorbitan monoleate (Tween 80) was used, 1% of which was incorporated in isotonic salt solution, distilled water and T.M.B. It was noted that the rough variants emulsi-

fied in Tween solutions gave very smooth homogeneous mixtures. Tests were put up using suspensions of the rough variant alone and also mixed equally with E. coli. With the later suspension however "blocking" was still found and the R(TY2) could not pass into the paper. As will be seen from Table 4, the organism alone passed freely into the filter paper for several centimetres.

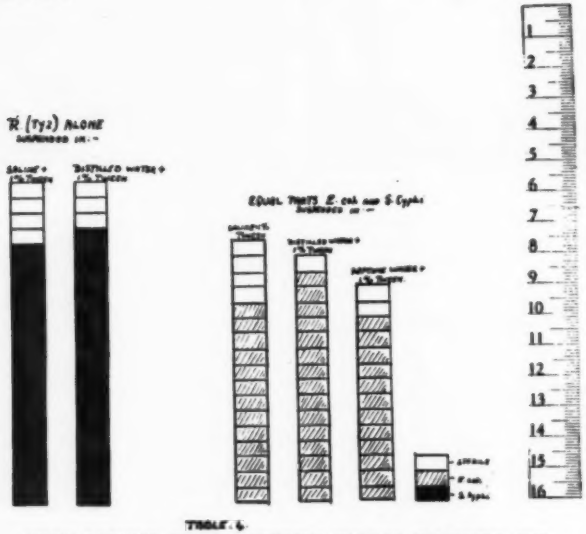


TABLE 4.

ISOLATION OF E. TYPHI FROM FAECES BY FILTER PAPER METHOD

A sample of faeces was obtained from a patient suffering with a chronic typhoid infection, for the purpose of trying the filter paper method as an aid in the recovery of the specific organism. It is difficult to assess the value of this test as with the present day methods in which highly selective media, and specific techniques are employed, the chances of being unable to recover the specific organism are extremely remote, even though such organisms be scantily excreted.

The specimen was examined by both the filter paper method and in parallel with various selective mediums. The organism was recovered equally well by both methods. A similar observation was recorded by Colquhoun in his paper in 1930 using the methods that were in vogue at that time. A brief mention is made of the fact that faecal saline suspensions, irrespective of whether these be concentrated or fairly dilute, do not readily

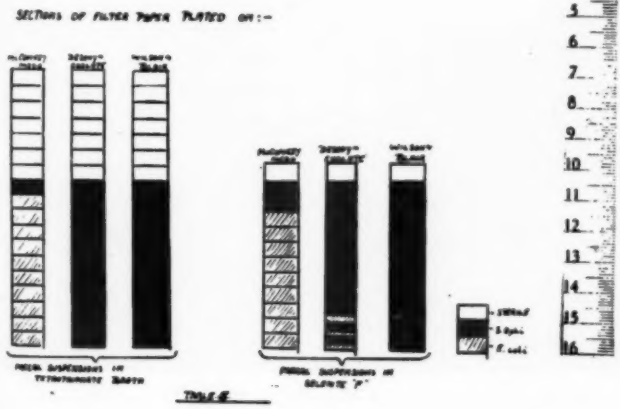
pass into the filter paper, this is probably due to the viscosity of the faecal sample. This can be overcome by inoculating a tube of peptone water with a particle of faeces the size of a cherry stone, incubating for four hours, after which the filter paper is suspended directly into the peptone water culture. Sections of the paper give a good recovery by the routine method described. In view of this finding, it was thought possible to improve the method using faecal suspensions suspended in enrichment fluids, which normally enhance the recovery of the typhoid organism by the usual laboratory method. Dilutions of the faeces therefore were made in:

(a) Tetrothionate Broth
(b) Selenite F.

Standard filter papers were suspended in each of these fluids for 40 mins. after which ½ cm. sections were transferred to tubes of peptone water, which were incubated for 18 hours. Subcultures were then made onto McConkey, Wilson and Blair and Desoxycholate media. As will be seen from Table 5, excellent results were obtained from the latter two mediums, due presumably to their normal selective action. The sub-cultures onto the McConkey media gave in comparison a very indifferent yield.

The results obtained from the direct platings of the specimen gave similar results to above, viz: Good recovery with Desoxycholate and W. and B., but McConkeys very poor in comparison.

Colquhoun reported that recovery of S. typhi in a dilution of 1-2000 was possible by the filter



Photograph below shows the ½ cm. sections after transfer to peptone-water and incubated for 18 hours. The tubes shown contain the top 3 cm. of the wetted strip. Note tubes 1 and 2, which are sterile; Nos. 2-4, differences in the degree of turbidity, these being pure cultures of S. typhi. Nos. 5-6 contain practically pure culture E. coli. Occasional colonies only of S (TY2).

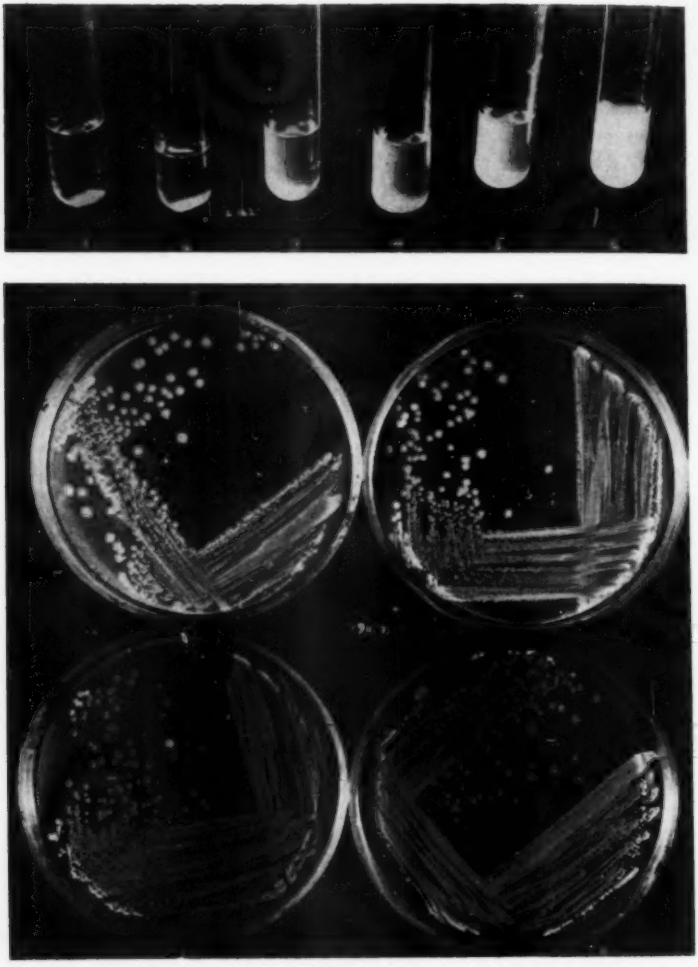


Plate cultures made from tubes 2-6, shown below. Top two plates being pure cultures of S. typhi (tubes 3-4). Lower two plates, pure cultures E. coli except for occasional colony of S. typhi, Tubes 5-6.

Fig. 3

paper method. Using equal parts of S. typhi and E. coli standardized to opacity 3, a range of ten fold dilutions was prepared. On testing these serial dilutions, clear cut recovery of S(TY2) was obtained up to dilutions of 1-1000. No typhoid organisms could be found by plating out samples of the 1-2000 dilution. Pure cultures of E. coli alone were obtained from each of the levels tested.

A dilution of 1 in 1500 was tested, and while this showed a few colonies of S (TY2) at the higher levels, the same clear cut pure cultures as obtained at dilutions of 1-1000 and in more concentrated solutions was not obtained.

SUMMARY

A simple method of isolating S. typhi is described which is dependent on the differences of the electric charge carried by bacteria. Utilizing strips of filter paper dipped into mixtures of equal parts of E. coli and S. typhi for 40 minutes pure cultures of S. typhi are recovered at certain levels, from the wetted strip. Below this level E. coli will be found almost in pure culture, but showing an occasional colony of S. typhi.

Bibulous paper being negatively charged will allow only particles with a like charge to pass into it. Both S. typhi and E. coli must therefore carry the negative charge, but the former organism is always found to go to a higher level than the latter. Samples of S(TY2) diluted 1-1000 gave pure plate cultures from the top of the strip. From higher dilutions however the typhoid organisms could not be recovered.

Faecal suspensions were treated similarly. It was difficult to assess the value of the filter paper method in the isolation of S. typhi, only one sample of faeces was examined, this being obtained from a typhoid carrier. Direct platings by the modern methods used for isolation of such organisms gave excellent recovery of the specific organism. The only apparent advantage would seem to be that with the aid of the filter paper, pure plate cultures of S. typhi could be obtained with the complete exclusion of E. coli. A marked zone occurs at regular levels between these organisms.

Rough strains of TY2 when mixed with E. coli will not pass into the paper. Mixtures of these two organisms seem to produce a blocking effect. The rough strain, which was markedly hyperagglutinable, when suspended alone in isotonic saline and in distilled water, will however pass freely into the paper.

Acknowledgment is made to the Chief Scientist, British Ministry of Supply, for permission to publish.

REFERENCES

1. (1929) Bedson, S. P., and Bland, J. O. W., Brit. Journ. Exper. Path., Vol. 10, p. 67.
2. (1930) Colquhoun, D. B., Lab. Journ. Vol. 6, No. 12, p. 381.

IN VITRO SUSCEPTIBILITY OF PATHOGENIC STRAINS OF *PSEUDOMONAS AERUGINOSA* AND *PROTEUS VULGARIS* TO THE NEWER CHEMOTHERAPEUTIC AGENTS.[2]

BERKLEY H. JOHNSON, M.S.[1]

There is a tendency for clinicians to employ antibiotics and other chemotherapeutic agents without regard to in vitro sensitivity tests at the beginning of treatment and then retesting during the longer periods in some cases that may be necessary for a clinicial response. Antimicrobial therapy, especially against such organisms as *Pseudomonas aeruginosa* and *Proteus vulgaris* many times will be disappointing without the performance of sensitivity tests. There is a close correlation between in vitro sensitivity tests of the infecting organisms and the clinical responses to any antibiotic tested.[1,2]

This procedure identifies the drug fast strains within the same species. There is a tendency among commercial producers of these agents to give a wide bacterial spectrum for their products without mentioning the genetic mutations and biochemical specificity variations between organisms in the same genus. Such factors as natural occuring resistant strains, acquired drug resistance and induced resistance should be evaluated by sensitivity testing.

This technique should be adopted both in mixed and pure cultures as isolated in the pathological condition. Many times in mixed infections after treatment there may still remain a resistant surviving population of the infecting organisms. When sensitivity tests are performed prior to and during treatment, it also gives the key for using the synergistic mixtures as reported in the many articles now appearing on the efficacy of combined therapy.[3,4,5] This is especially true when gram positive organisms are mixed with a gram negative flora.

DISCUSSION

Interest in *Pseudomonas aeruginosa* and *Proteus vulgaris* was aroused by their frequent and wide distribution in chronic infections of various kinds, such as, urinary, otitis media, wounds and a case of gastroenterocolitis *(Ps. aeruginosa)*. The fact that they are

[1] Bacteriologist, Veterans Administration Hospital, Walla Walla, Washington.
[2] Reviewed in the Veterans Administration and published with the approval of the Acting Chief Medical Director. The statements and conclusions published by the author are the result of his own study and do not necessarily reflect the opinion or policy of the Veterans Administration.

quite (innately) resistant or rapidly acquire resistance to many of the common chemotherapeutic agents has been observed. Variability is reflected in these organisms as some strains are moderately susceptible to sulfonamides, others to streptomycin, chloromycetin, aureomycin and terramycin.

The chemotherapeutic agents used in this study were terramycin, streptomycin, dihydrostreptomycin, aureomycin, bacitracin, chlormycetin, gantrisin and a triple sulfonamide mixture. The method for testing the sensitivity of the bacterial strain to these antimicrobial agents was by the paper disk technique. The disks being impregnated with varying concentrations of the agent to be tested.[5] This technique is simple and seems quite sufficient for practical evaluations of sensitivities as applied clinically. Tube methods and subcultures for bacteriostatic studies were used when indicated. The results have been obtained over a period of one year with different types of infections by these organisms. Several cases of each type have been studied although a limited number of cases were available at our hospital as it is primarily a tuberculosis unit.

The real purpose of this article, however, is to suggest the importance of dealing with each organism of this group as a separate sensitivity problem. A scheme derived from previous performance of sensitivity is useful as a guide, but certainly has limited applications. The following chemotherapeutic agents of choice are suggested in the order of their in vitro efficacy for testing this series of organisms as isolated:

ORGANISM TESTED	DRUG OF CHOICE
A. *Proteus vulgaris*	1. Chloromycetin
	2. Streptomycin or Dihydrostreptomycin
	3. Gantrisin or a triple sulfonamide mixture.
B. *Pseudomonas aeruginosa*	1. Terramycin
	2. Streptomycin or Dihydrostreptomycin
	3. Aureomycin

The source of the culture as to the site of infection, of course, has no influence on the primary susceptibility of a strain tested. Many failures in treatment are certainly associated with the pathological condition, character of the lesion and other clinical aspects. The pathological condition being favorable for chemotherapeutic therapy, failures in the clinical response may then be due to acquired resistance to the test chemotherapeutic agent. The second choice of the antimicrobial agents tested is

then recommended, etc. This presents a logical approach to treatment. Each organism is isolated and dealt with separately, which is reflected by a savings in time and financial costs to the patient. This technique aids in eliminating the trial and error method in chemotherapeutic treatment. This is especially true when other factors are favorable for basic chemotherapeutic treatment.

CONCLUSIONS

1. There is a definite correlation between in vitro susceptibility of these organisms and their response to antimicrobial agents.

2. The drugs of choice for these organisms are listed (with reservations) in sequence of their efficacy as observed on a limited number of cultures.

3. An attempt has been made toward specific therapy with respect to the results obtained from each culture, then following extended cases by further in vitro susceptibility studies whenever deemed necessary by the clinician.

4. The ideal and rational approach is to deal with each organism, as such, especially if the clinical response is unfavorable and the associated pathological condition has been corrected which might limit the effectiveness of chemotherapeutic therapy.

REFERENCES

1. Taylor, R. D.: Postgrad Med. 7:216 (March) 1950.
2. Mulholland, S. W.: Pennsylvania M. J. 53:240 March 1950.
3. Schmelkes, F. C. and Wyss, O.: Proc. Soc. Exptl. Biol. Med. 49:263-267.
4. Spicer, Sophie: The J. of Lab. Clinical Med., Vol. 36, No. 2, Pages 183-191, August 1950.
5. Klein, M., and Kimmelman, L. J.: J. Bact. 54:367, 1947.
6. Spicer, Sophie and Blitz, Diana: J. Lab. and Clin. Med. 33:417, 1948.

THE GAVEL

Last call for 1951 Convention! As the time grows nearer and more information reaches you, I know that more and more of you are finding that call as irresistible as the will-o-the-wisp. Unlike that light, however, the rays of convention publicity are easily traced to their source; and their source looks from this distance like the brightest star yet in our galaxy of brilliant conventions. We are truly looking forward to seeing all of you who have been attending conventions, plus many more, in Swampscott, June 24-28. Just don't disappoint us, and we won't disappoint you.

Elsewhere in this Journal and the past one, you will find all the information about the scientific and entertainment aspects of the program; here we give you what information we have at this point on the business to be transacted. Let me remind you again that the reports of the Nominations and Elections Committee and the Constitution and By-Laws Committee, published in the March Journal, should be carefully studied and all delegates instructed as to the wishes of the Society which they represent; I cannot over-stress the importance of considered deliberation at home on the questions to be acted upon in the House of Delegates. Under Old Business, a report from a committee from the Board will be heard giving the results of their investigation concerning our official seal and the one proposed last year to replace it. We are sure that many of you have also given this matter earnest consideration, and your conclusions will be welcomed in the discussion before the House takes final action. Under New Business will be the presentation of the budget for 1951-52 for adoption. The Finance Committee is now working on that budget, and if any of you have suggestions concerning it now is the time to communicate with the committee; they will welcome your ideas and contributions, and receiving them now will prevent loss of time in revision at the time of the meeting.

Each standing committee of ASMT will present a work-shop this June at the Convention, and they will be just as the name implies: meetings at which the national committee will guide the discussion with the aim of working out problems peculiar to the functioning of that particular committee and cooperation between the committee and its counterparts in the state societies. I strongly urge that, as far as possible, all state committee chairmen attend the work-shop for their committees; the mutual benefit will increase in direct proportion to the percentage of complete representation.

The Advisory Council will meet, as usual, prior to the House of Delegates; if your state president cannot attend, please see that you have a representative at the Council meeting. This meeting is for the purpose of discussing organizational problems and is a vital element in the successful progress made by ASTM and its affiliated societies. Although it is advisory in nature, and cannot therefore make official decisions, it can and does make recommendations to the Board of Directors and through them to the House of Delegates. Those recommendations are given careful consideration and carry much weight; it is a sort of unofficial 'Senate,' so you state presidents be sure to be there or send an alternate—and come prepared to take full advantage of the opportunity to be heard if you have any progressive ideas or want advice from other groups.

I am looking forward to seeing you in Swampscott; do come. In the meantime, I will be happy to hear from you if you have any suggestions, problems, or gripes, but most especially if you have suggestions!

V. J. S.

CLINICAL BIOCHEMISTRY

Refresher Course

July 9-14, 1951, Inclusive
(9-12 noon, 1-5 P.M. daily)

To be presented at Baylor University College of Medicine, Houston, Texas, by the Department of Biochemistry
Dr. Joseph H. Gast, Professor and Chairman
Dr. Ferrin B. Moreland, Associate Professor
Dr. Russel O. Bowman, Associate Professor
Consulting biochemists to Hermann, Houston Negro, Jefferson Davis, Methodist, San Jacinto Memorial, Southern Pacific and Veterans Administration Hospitals.

PLAN OF COURSE

(Lectures, laboratory, conferences and demonstrations)

1. Review Fundamental Principles of Quantitative Chemistry
   Use of balance
   Glassware calibration
   Standard solutions
   Reagents
   Buffers
   Chemical arithmetic
   Titrations
2. Colorimetry and Spectrophotometry
   Standardization: use of factors, tables, curves and graphs, blanks, controls, standards and replicates
   Detection of mechanical errors
3. Selection and Evaluation of Procedures
   Simplicity vs. Accuracy, Specificity, Elimination of unnecessary duplication.
4. Preparation of Patient—Collection and Handling of Specimens, Preservatives, etc.
5. Special Equipment and Techniques
   pH meters, flame photometer, fluorophotometer, micro methods.
   Gasometric methods.
6. Trouble Shooting
   Finding and correcting errors. Do's and Don't's.

Make application to Dr. J. H. Gast, Department of Biochemistry, Baylor University College of Medicine, Houston 5, Texas.
Limited to 60 registered M.T. (ASCP). Give your registry number.
Last date for registration—June 20, 1951.
Fee: $25—$10 deposit with application. No refunds after June 25, 1951.
Requests for room reservations can be made with applications.
Rates will be as follows:
Shamrock Hotel—Air Conditioned
$12 per day—2, 3 or 4 persons per room (single beds)
Air Conditioned Tourist Courts
Single—$24-$30 per week. Double—$28-$35 per week.
State number or names of persons you wish to share room.

UNESCO GIFT COUPON PLAN

Many requests have come from state societies for information regarding the shipment of used equipment, glassware and such. The answer, just in case your society is thinking in that way, is that such things are too expensive to pack and ship. The same amount of money would go much farther.

Now we have information on the UNESCO GIFT COUPON PLAN. This is a device to make it easy for individuals through organizations to contribute to Educationa! Reconstruction projects. By buying stamps at 25 cents each, individuals may contribute to the purchase of Gift Coupons in ten-dollar denominations to be sent to educational institutions in war-devastated countries. These institutions use the coupons in the regular commercial markets of the world as an international money order to obtain a wide range of educational materials.

UNESCO maintains priority lists of institutions in the war-devastated and under-developed countries together with lists of their specific needs. From these lists organizations may select projects around which to plan their own part of the Gift Coupon program.

If your society can give as much as $10.00 you may write directly for booklets; if not, contribute to the ASMT general fund. When you write, mention that your group is an affiliate of ASMT. Write to: UNESCO Reconstruction Liaison Office, Room 2201, Secretariat Bldg., United Nations, N. Y. Remember to report to the Public Relations Committee what contributions you have made.

Doris E. Boon, M.T. (ASCP)
Public Relations Committee
1588 Ansel Road
Cleveland 6, Ohio

CANADA

The national convention of the Canadian Society of Laboratory Technologists will be held in Winnipeg, Manitoba, Canada, at the Fort Garry Hotel from June 24th to June 27th inclusive. A fine program of scientific papers and technical exhibits has been planned. The entertainment will be varied and interesting. It is hoped that members of the American Society of Laboratory Technologists will be able to attend.

More detailed information can be obtained by writing:

Miss Miriam Wiseman,
Municipal Hospitals,
Winnipeg, Manitoba, Canada.

COMBINE YOUR VACATION WITH CONVENTION TIME

Miss Marion Alcott suggests that there are day trips to Provincetown on Cape Cod or Nantucket, two or three day trips to the White Mountains, also to Montreal, Quebec, or Nova Scotia. Any of these can be fitted to your time pre- or post-convention. Consult your local travel agency.

KNOW YOUR CONVENTION HOST—Massachusetts Association of Medical Technologists

Beside the many duties contingent with the organization of an over-all convention program, our host for the 1951 meeting, the Massachusetts Association of Medical Technologists has carried on a full (and then some) quota program of participation in other activities. The NEW ENGLAND JOURNAL of MEDICINE for February 8, 1951, carried the Symposium on "What about Medical Technologists," with "The Problem of the Medical Technologist," by Robert A. MacCready, M.D., "Educational Requirements of the Medical Technologist," by Philip M. Richardson, Ph.D., "Training of the Medical Technologist," by Elinor Judd, A.B., M.T. (ASCP), and "Problems in the Maintenance of Adequate Laboratory Staffs," by Donald A. Nickerson, M.D. This panel had been presented at the annual meeting of the Massachusetts Medical Society in May, 1950, and at a meeting of the Mass. Hospital Association in January, 1951. This society does not stop at an **annual** convention, but has from two to three other meetings each year. These are held in different parts of the state to make it possible for all members to attend. Their programs are of the same high calibre as the ones presented before our national group. Plans are to consider the possibility of a series of Seminars to be held over the state. Miss Geneva Daland was elected an honorary member of the state society.

In Civil Defense planning the Association has taken the lead in following the recommendations of the New England Blood Bank Association and has made an excellent start toward enrolling all possible personnel in the laboratory section of this effort. The Membership Committee is responsible for the compilation of a Roster of all available laboratory workers. A placement service has been maintained with the same results as in other parts of the country: there are still too few qualified medical technologists to fill all needs.

Through its Public Relations Committee contacts have been made with related Medical organizations. This committee has also cooperated with the ASMT P.R. Committee in all the projects recommended by the national society.

The Research Committee has developed a project which could well be emulated by other societies who wonder what functions this committee has; from their report, "A list of titles and reprints which have been collected by the Worcester District MT Association was sent to each local group, with the suggestion that they start a collection of reprints for their own district. Where reprints cannot be duplicated, they can be exchanged."

The Education Committee made contacts with all High Schools and Colleges within the state, through vocational guidance counselors, with resulting talks before groups on Career Days and the distribution of information on medical technology. Numerous short courses have been presented by various colleges and universities, and several of these schools have been approached on the ubject of offering degrees in Medical Technology. The Association is raising funds for the production of a sound and color film on Medical Technology. The N.E. Pathologists Society, as well as local groups and individuals are participating in this effort.

The Standards and Studies Committee has conducted a survey of conditions in medical technology in Massachusetts.

Upon the completion of their state meeting in Springfield, on April 21, the members returned to their respective homes to take part in the National Laboratory "Open House" on the following day. There were at least two or three hospital laboratories participating in each district.

LET'S ALL GO TO THE CONVENTION IN SWAMPSCOTT, MASSACHUSETTS, AND SEE HOW THEY'VE ACCOMPLISHED ALL THIS WORK. WE'LL SEE YOU THERE JUNE 24-28, 1951.

CONVENTION ANNOUNCEMENTS

Serving as Honorary Chairman for the 19th Annual Convention of ASMT will be John F. Conlin, M.D., Director of Medical Information and Education for the Massachusetts Medical Society. Dr. Conlin is an ardent supporter of Medical Technologists.

Bring your copy of the March Journal to the meeting so that you will have a copy of the proposed amendments to the Constitution and By-Laws.

The Registration and Credentials Desk will open at 8:00 A.M. Sunday, June 24. Hostesses will be able to direct you to the church of your choice. Sunday afternoon will feature tours of historic Boston, leaving the New Ocean House at 2:00 P.M. for the tour of Lexington and Concord, and at 3:00 P.M. for the tour of Boston. Plan to arrive early on Sunday so that you will be able to visit these historic spots that have been so important in our country's development.

Advisory Council Meeting at 2:00 P.M.

All the New England State Associations will be co-hostesses at the Reception Sunday evening from 8:00 to 10:00 P.M. in the Ball Room. This is your opportunity to meet the Candidates for election. Talk to them, get their viewpoints, and tell them yours.

For merriment and fun, there will be a Country Dance Monday evening, June 25. If you prefer, come and show your skill at cards.

Tuesday, June 26, at 8:00 P.M., Verne Williams of WBZ Boston will be the Master of Ceremonies at a Quiz Program. Perhaps you will be one of the lucky ten picked from the audience for participation and be eligible for prizes.

A trip to the famous fishing port of Gloucester is a must for all visitors to New England. Leaving the New Ocean House at 2:00 P.M., on Wednesday, June 27, you will go one way by sea, and the other by land. See the statue of the Gloucester Fisherman where the famous memorial ceremonies are held for the fishermen lost at sea. A New England Clam Bake, known the world over for its excellence, will be waiting for you when you get back to the New Ocean House.

Thursday evening, June 28, the annual banquet will be held at 7:00 P.M., at the New Ocean House. Convention Awards will be made. John Nicol Mark, Scotch Humorist, will be the guest speaker with a flavor of New England.

DON'T FORGET TO BRING:

1. Your bathing suit as you'll want a dip in the refreshing Atlantic Ocean
2. Your golf clubs
3. Your tennis racquet
4. Last but not least a sweater, for as Mark Twain once said, "If you don't like the New England weather—wait a minute."

Please don't send any money in advance. If you do, refunds will be made after the Convention.

Make your reservations today for the hotel with Miss Mary Duffy, Assistant Manager, New Ocean House, Swampscott, Massachusetts. See the March Journal for full details.

If possible, notify Miss J. Clare MacDonald, Robert B. Brigham Hospital, Boston, Mass., of the time of your arrival in Boston, so that bus transportation to Swampscott may be arranged. Otherwise it will be necessary to transfer from the South Station to the North Station to take a train to the convention place. From Chicago, Detroit, Toledo, Cleveland, Toronto, Hamilton, Buffalo, Albany, trains will arrive in Boston: A.M., 7:10, 9:05, 9:35, 10:30. P.M., 12:10, 2:40, 8:45.

From St. Louis, Indianapolis, Cincinnati, Dayton, Columbus: A.M. 7:10, 9:05, 10:30; P.M. 2:40.

BE SURE TO HAVE YOUR ASMT MEMBERSHIP CARD WHEN YOU REGISTER FOR IDENTIFICATION FOR HOUSE OF DELEGATES!

Entertainment

To facilitate the planning of the Entertainment, please check those functions you wish to attend and send them to: Miss Marion D. Alcott, 64 Lincoln Street, Watertown, Massachusetts.

Sunday, June 24:

2:00 P.M. Lexington and Concord sightseeing trip_____
or
3:00 P.M. Historic Boston sightseeing trip_____

8:00 P.M. Reception _____

Monday, June 25:

8:30 P.M. Country Dance or Cards_____

Tuesday, June 26:

8:30 P.M. "Cinderella Weekend" Quiz Program, with Verne Williams as M. C._____

Wednesday, June 27:

1:00 P.M. Boat and bus trip to Gloucester_____

Thursday, June 28:

7:00 P.M. Banquet—Guest Speaker—
John Nicol Mark, Scotch Humorist_____

Total cost for all social events $13.00 or $15.00 depending upon which tour you select. Individual day registration and entertainment may be arranged.

ASMT members must have their membership cards for identification in order to attend House of Delegates meeting.

Hotel Arrangements for June 24-28, 1951

Make your reservations directly with the New Ocean House, Swampscott, Massachusetts. Please give the following information: Name, Address, Date of arrival, Time, Date of departure, Time, Accommodations desired. Designate time desired for American Society of Medical Technologists convention.

RATES	Daily per person including Room and Meals
Double Room with Bath, Twin Beds	$11.00 and $11.50
Double Room with Bath, Twin Beds, Hotel Cottages	10.00 and 11.50
Large Room with Bath, Three Beds	9.50 and 10.00
Large Room with Bath, Four Beds	9.00 and 9.25
Double Room with Running Water, Twin Beds	9.50 and 10.50
Suite, 2 Double Rooms, Bath between, Four Beds	9.75 and 10.50
Suite, 2 Double Rooms, Bath between, Six Beds	9.25 and 9.50
Choice Corner or Bay-Window Room, Bath, Twin Beds	12.00 and 13.00
Single Room with Bath	13.00 and 14.00
Single Room with Bath, Hotel Cottages	12.00 and 12.50

If a sufficient number of Sisters wish hotel accommodations, a separate block of rooms will be reserved for them. If need be, a separate cottage will be reserved for them. For Sisters Reservations, write Sister Mary Claire, 415 West 51st Street, New York, New York.

If you find that you must cancel your reservations, write to Miss Mary Duffy, Assistant Manager, New Ocean House, Swampscott, Massachusetts.

Make Your Reservations Today

ASMT CONVENTION

New England and Massachusetts open their arms to you June 24-28, 1951 at YOUR National Convention, New Ocean House, Swampscott, Massachusetts. We shall have excellent papers, fun and friendliness to offer. Come and tell us about the AMA SPONSORED SOCIETY. Let us hear top ASMT PAPERS. Let us join hands in MEDICAL TECHNOLOGY! COME!

Mrs. Elinor Judd, M.T. (ASCP)
General Chairman 1951 National Convention

THE AMERICAN SOCIETY OF MEDICAL TECHNOLOGISTS

NINETEENTH ANNUAL CONVENTION

Headquarters
NEW OCEAN HOUSE
SWAMPSCOTT, MASSACHUSETTS
June 24, 25, 26, 27 and 28, 1951

Program

SUNDAY, JUNE 24

8:00 A.M. Registration Opens
2:00 P.M. Advisory Council Meeting—Ballroom—New Ocean House
8:00 to 10:00 P.M. Reception—Ballroom—New Ocean House

MONDAY, JUNE 25—Convention Hall

8:00 A.M. Formal Opening of Technical and Scientific Exhibits.
9:00 A.M. National Anthem: Mrs. Lois A. Nellis
Invocation:
The Right Reverend Norman B. Nash, D.D., the Episcopal Bishop of Massachusetts.
Welcome:
Lt. Thomas F. Casey, USNR, Military Staff of Governor Paul A. Dever
Andrew R. Linscott, Selectman, Swampscott, Massachusetts
Greetings:
Vernal J. Schene, M.T. (ASCP), President, American Society of Medical Technologists
ASMT membership cards must be presented at registration time in order to attend meeting of House of Delegates.
10:00 A.M. House of Delegates Meeting—Convention Hall
12:30 Noon Recess
2:00 P.M. House of Delegates Reconvenes

TUESDAY, JUNE 26—Convention Hall

8:00 A.M. Exhibits Open
8:00 A.M. "Report on the Board of Registry of Medical Technologists," Lall G. Montgomery, M.D., Chairman, Registry of Medical Technologists, Muncie, Indiana
8:30 A.M. "A Specific Hemagglutination Test in the Diagnosis of Tuberculosis." Frances G. Murry, M.T. (ASCP), St. Louis, Missouri
9:00 A.M. Symposium: "The Role of the Medical Technologist in an Atomic Disaster." Representatives of the Biological and Medical Research Division, Argonne National Laboratory; Depart-

ment of Medicine, University of Chicago, Chicago, Illinois, and Harvard Medical School, Boston, Massaschusetts

1. General Remarks on the Professional Problem Involved.
   Ellen Skirmont, M.T. (ASCP), Argonne National Laboratory
2. Survey Hematologic Studies and Their Significance
   Evelyn Gaston, Argonne National Laboratory
3. Special Studies on Blood Coagulation
   Margaret Sanderson, M.T. (ASCP), Argonne National Laboratory
4. Bacteriologic Problems
   Isabelle Havens, M.T. (ASCP), University of Chicago, Department of Medicine
5. Present State of Knowledge of Formed Elements of the Blood: Preservation and Potential Use
   a. Leucocyte Studies
      Cathleen Alban, M.T. (ASCP), Harvard Medical School
   b. Platelet Studies
      Grace Kemp, Harvard Medical School
   c. Efforts to Isolate and Stabilize Prothrombin
      John McHugh, Harvard Medical School
6. Summary and Discussion
   Leon O. Jacobson, M.D., Associate Professor of Medicine and Associate Dean of Biological Science Division University of Chicago, Albert Merritt Billings Hospital and Consultant to the Biology and Medical Division of the Argonne National Laboratory

10:00 A.M. VISIT EXHIBITS

10:30 A.M. "Atomic Bomb Radiation Casualties: Their Diagnosis, Treatment and Prognosis." Dudley P. Jackson, Lt. j.g. M.C.R. USNR, Naval Medical Research Institute, National Naval Medical Center, Bethesda, Maryland

11:00 A.M. "Blood Groups and Blood Group Antibodies." Louis K. Diamond, M.D., Associate Professor of Pediatrics, Harvard Medical School, Director of Hematology Research Laboratory, Director of the Blood Grouping Laboratory, Children's Medical Center, Boston, Massachusetts

11:30 A.M. "Homologous Serum Hepatitis: An Occupational Hazard for the Medical Technologist." Sydney S. Gellis, M.D., Physician, Children's Medical Center, Boston, Massachusetts, Pediatrician-in-Chief, Beth Israel Hospital, Boston, Massachusetts, Assistant Professor in Pediatrics, Harvard Medical School, Consultant in Pediatrics to the Joslin Clinic

12:00 Noon "What Can We Learn from a Blood Film?" Geneva A. Daland, B.S., Research Laboratory Technician in Hematology, Thorndike Memorial Laboratory, Boston City Hospital, Boston Massachusetts.

12:30 Noon Recess

TUESDAY, JUNE 26, P.M., Convention Hall

1:30 P.M. "Studies Based on Errors Observed in the Use of Anticoagulants in Blood Chemistry." Mary Benedict Clark, M.T. (ASCP), Louisville, Kentucky

2:00 P.M. "Dubos' Solid Agar Medium in Routine Laboratory Diagnosis of Tuberculosis." Mary E. Molloy, M.T. (ASCP), Philip B. Miner, B.S., M.S., and Roland E. Berry, A.B.

2:30 P.M. "Use of Blood and Blood Derivatives." Charles A. Janeway, M.D., Thomas Morgan Rotch Professor of Pediatrics, Harvard Medical School, Physician-in-Chief, Children's Medical Center, Boston, Mass.

3:00 P.M. "Diagnosis of Hemolytic Anemias." Frank H. Gardner, M.D., Instructor in Medicine, Harvard Medical School, Boston, Massachusetts, Associate in Medicine, Peter B. Brigham Hospital, Boston, Massachusetts, Associate Hematologist, Boston Lying-in Hospital, Boston, Massachusetts
3:30 P.M. VISIT EXHIBITS
4:00 P.M. "Changes Found in the Blood and Hematopoietic Function in Chronic Lead Exposed Painters." Wilhelmina Schroeder, M.T. (ASCP), Baytown, Texas
4:15 P.M. "Laboratory Planning"—New Hampshire Society of Medical Technologists, presented by Sister Mary of Mercy, J. O. M. Sacred Heart Hospital, Manchester, New Hampshire
4:30 P.M. Workshop: Membership—Presiding, Ida L. Reilly, M.T. (ASCP), assisted by Committee Members

WEDNESDAY, JUNE 27

Section One: Convention Hall

8:00 A.M. Exhibits Open
8:00 A.M. "A New Method for Detection of Albumin in Urine." Phyllis Stanley, M.T. (ASCP), Newark, New Jersey
8:30 A.M. "Uses and Abuses of the Flame Photometer." Anne J. Sommer, M.T. (ASCP), St. Louis, Missouri
9:00 A.M. "The Registered Medical Technologist: Her Position in the Physician's Office." Rebecca Irving, M.T. (ASCP), Quincy, Massachusetts
9:30 A.M. "Methods of Isolation and Incidence of Brucella Types Found in Georgia." Alice Brim, M.T. (ASCP), Georgia Department of Public Health, Atlanta, Georgia
10:00 A.M. VISIT EXHIBITS
10:30 A.M. "The Present Status of Cortisone and ACTH." Theodore B. Bayles, M.D., Clinical Associate in Medicine, Harvard Medical School, Associate in Medicine, Peter B. Brigham Hospital, Boston, Massachusetts, Research Director, Robert B. Brigham Hospital, Boston, Massachusetts
11:00 A.M. "The Importance of the Laboratory in the Management of Diabetes." Priscilla White, M.D., Physician, New England Deaconess Hospital, Boston, Massachusetts, Instructor in Pediatrics, Tufts Medical School, Boston, Massachusetts
11:30 A.M. "Sensitivity of Bacteria to Antibiotics." George G. Jackson, M.D., Milton Fellow, Harvard Medical School, Boston, Massachusetts, Research Fellow, Thorndike Memorial Laboratory, Boston City Hospital, Boston, Massachusetts, Assistant in Medicine, Boston City Hospital, Boston, Massachusetts
12:00 Noon "Two Clinical Procedures of Interest to Medical Technologists: 1. Serum Bilirubin. 2. Lipid Phosphorus with Adaptation to the Coleman Spectrophotometer." Elizabeth Maclay, M.T. (ASCP), Rochester, Minnesota
12:30 Noon Recess

Section Two: Ballroom

8:00 A.M. Workshop: Recruitment and Vocational Guidance—Presiding, Ruth Feucht, M.T. (ASCP), assisted by Regina Duane, M.T. (ASCP), Bennie Bargas, M.T. (ASCP), Everlyn Jardine, M.T. (ASCP), Edna Rigdon, M.T. (ASCP), and Oscar M. Alton, M.T. (ASCP).
9:00 A.M. Workshop: Public Relations and Publicity—Presiding, Frieda Claussen, M.T. (ASCP), assisted by Lavina White, M.T.

(ASCP), Mary Nix, M. T. (ASCP), Doris Boon, M.T. (ASCP), Josephine Pyle, M.T. (ASCP), and Marion Baker, M.T. (ASCP).

1:30 P.M. See Entertainment

6:30 P.M. Priscilla Room: Alpha Mu Tau Dinner Meeting

8:00 P.M. Workshop: Legislation—Ballroom—Presiding, Sadie Cartwright, M.T. (ASCP), assisted by Dorothy Lee, M.T. (ASCP), Sr. Mary Clare, O.S.F., Dorothy Prest, M.T. (ASCP), Dorothy Crane, M.T. (ASCP), Barbara Isbell, M.T. (ASCP), Virginia Burris, M.T. (ASCP), Grace Mary Ederer, M.T. (ASCP), and Carolyn Martin, M.T. (ASCP).

THURSDAY, JUNE 28

Section One: Convention Hall

8:00 A.M. Exhibits Open

8:00 A.M. "About Medical Technology—A Survey." Sister Mary Alcuin, O.S.B., M.T. (ASCP), and Students in Medical Technology, College of St. Scholastica, Duluth, Minnesota

8:30 A.M. "Cultural Diagnosis of Brucellosis." Mabel V. Meeks, M.T. (ASCP), Newington, Connecticut

9:00 A.M. "Micro Technique for Prothrombin Times." Jeanette Sanduzzi, M.T. (ASCP), Montclair, New Jersey

9:15 A.M. "Experiences with Rh Sensitivity Titres." Lydia Brownhill, M.T. (ASCP), Meriden, Connecticut

9:30 A.M. "Diabetic Detection." Doris M. Wallace, M.T. (ASCP), Houston, Texas

9:50 A.M. "The Isolation of Pathogens in Suspected Food Poisoning." Adabelle Noyes, M.T. (ASCP), Fort Baker, California

10:10 A.M. VISIT EXHIBITS

10:30 A.M. "The Role of the Bacteriologist in the Chemotherapy of Infection." Louis Weinstein, M.D., Associate Professor of Medicine, Boston University School of Medicine, Lecturer in Infectious Diseases, Harvard Medical School, Chief of Service, Haynes Memorial Hospital, Massachusetts Memorial Hospitals, Boston, Massachusetts

11:00 A.M. "The Value of Serologic Tests for Syphilis." William A. Hinton, M.D., Professor Emeritus of Bacteriology and Immunology, Harvard Medical School, Physician-in-Chief of Clinical Laboratories, Boston Dispensary, Boston, Massachusetts, Chief, Massachusetts State Wassermann Laboratory

11:30 A.M. "Pulmonary Mycoses and the Diagnostic Laboratory." Leanor D. Haley, Ph.D., M.T. (ASCP), Yale University School of Medicine, New Haven, Connecticut

12:00 Noon "Bone Marrow in Health and Disease." Margaret A. Anderson, M.T. (ASCP), Boston, Massachusetts

12:30 Noon Recess

Section Two: Ball Room

Panel: "The Training of Medical Technologists"

8:00 A.M. Workshop: Standards and Studies. Presiding, Sister Mary Dolorosa, M.T. (ASCP), assisted by Sister Mary Leo Rita, S.S.M., M.T. (ASCP), Carolyn Martin, M.T. (ASCP), Elinor Judd, M.T. (ASCP), Agnes Marie Lyman, M.T. (ASCP), Anna Priest, M.T. (ASCP), and Hazel Current, M.T. (ASCP).

9:00 A.M. Workshop: Education—Presiding, Rose Hackman, M.T. (ASCP), assisted by Estelle Downer, M.T. (ASCP), Ruth Feucht, M.T. (ASCP), Elizabeth Frey, M.T. (ASCP), and Mary Frances James, M.T. (ASCP).

10:00 A.M. VISIT EXHIBITS

11:30 A.M. Workshop: Finance—Presiding, Mary Eichman, M.T. (ASCP), assisted by Rose Matthaei, M.T. (ASCP), Executive Secretary, Loretta Laughlin, M.T. (ASCP), Kathryn Dean, M.T. (ASCP), and Louise Vance, M.T. (ASCP).
12:30 Noon Recess

THURSDAY, JUNE 28, P.M.

1:30 P.M. "A Simple Screen Test for Cancer." Anna M. Slicher, M.T. (ASCP), Mount Vernon, New York
2:00 P.M. "Dubos' Solid Agar Medium in Routine Laboratory Diagnosis of Tuberculosis." Mary E. Mollay, M.T. (ASCP), Philip B. Miner, B.S., M.S., and Roland E. Berry, A.B.
2:30 P.M. "Fungi—Fact or Fancy for the Medical Technologist." Alice T. Marston, Ph.D., Assistant Professor of Microbiology, Boston University School of Medicine, Boston, Massachusetts
3:00 P.M. "Present Plans for the Commissioning of Clinical Laboratory Officers." O. F. Goriup, Colonel, M.S.C., Department of the Army, Office of the Surgeon-General, Washington 25, D. C.
3:30 P.M. Panel Discussion: "Medical Technologist in Civil Defense." Mollie Hill, M.T. (ASCP), North Arlington, Virginia
4:30 P.M. Workshop: Constitution and By-laws. Presiding, Lee M. Soucy, M.T. (ASCP), assisted by Committee Members.

PROGRAM—FIRST!

Entertainment Program

Sunday, June 24, 1951

2:00 P.M. Lexington and Concord sightseeing tour
or
3:00 P.M. Historic Boston sightseeing tour
8:00 P.M. Reception

Monday, June 25, 1951

8:00 P.M. Country Dance or Cards

Tuesday, June 26, 1951

8:00 P.M. "Cinderella Weekend" Quiz Program, with Verne Williams as M. C.

Wednesday, June 27, 1951

2:00 P.M. Boat and Bus Trip to Gloucester

Thursday, June 28, 1951

7:00 P.M. Annual Banquet: Awards—John Nicol Mark, Scotch Humorist, Guest Speaker

SCIENTIFIC EXHIBITS

1 Title The American Society of X-Ray Technicians
Sponsor: The American Society of X-Ray Technicians and The American Registry of X-Ray Technicians
2 Title Stars Fell on Alabama
Sponsor: The Alabama State Society of Medical Technologists
3 Title Frog (Rana Pipiens) Test for Pregnancy
Sponsor: Saint Mary's Hospital, Passaic, New Jersey
4 Title Lupus Erythematosis Phenomenon
Sponsor: Wayne University College of Medicine
5 Title Colorado State Society of Medical Technologists
Sponsor: Colorado State Society of Medical Technologists
6 Title Registry of Medical Technologists of the American Society of Clinical Pathologists
Sponsor: Lall G. Montgomery, M.D., Chairman, Board of Registry
7 Title New Hampshire Laboratory Planning—Hematology
Sponsor: New Hampshire Society of Medical Technologists

8 Title Ohio Society of Medical Technologists
Sponsor: Ohio Society of Medical Technologists
9 Title Laboratory Open House in Massachusetts
Sponsor: Massachusetts Association of Medical Technologists, Inc.
10 Title Growth and Activities of the Connecticut Society of Medical Technologists
Sponsor: Connecticut Society of Medical Technologists
11 Title Cleared Human Embryos in Plastic
Sponsor: Sarah O'Niell, MT (ASCP), Baylor University, Houston, Texas
12 Title Geneva Deland, Hematology
Sponsor: Massachusetts Association of Medical Technologists, Inc.
13 Title Method of Reporting Laboratory Findings to Doctors
Sponsor: Illinois Medical Technologists Association
14 Title Movie from Atomic Energy Commission showing effect of Radiation
Sponsor: Illinois Medical Technologists Association
15 Title 1952 Convention in Portland
Sponsor: Northwest Branch, Portland, Oregon
16 Title Exhibit of Large Bone and Whole Brain Sections
Sponsor: Dr. Gunn, Army Medical Illustration Service
17 Kentucky State Society of Medical Technologists

Technical Exhibitors and Booth Descriptions

LEDERLE LABORATORIES DIVISION
American Cyanamid Company
New York, N. Y.

You are cordially invited to visit our exhibit in Space 27, where you will find representatives who are prepared to give you the latest information on Lederle products.

MEINECKE & COMPANY, INC.
New York, N. Y.

Featuring Haemo-Sol, the chemically designed prepartion for the cleaning of laboratory apparatus with specific emphasis on laboratory glassware. One of the outstanding characteristics of Haemo-Sol is reflected by its complete solubility. In addition, it is fully Haemo-Digestive, creating a lower surface tension, and is compounded with a controlled pH. Haemo-Sol does not contain what is generally thought of as one, or more active ingredients, rather it is a fine adjustment of a mixed formula containing no caustics.

KIMBLE GLASS
Toledo, Ohio

Kimble Glass, Division of Owens-Illinois Glass Company, will exhibit a representative line of Kimble laboratory glassware, including volumetric and calibrated apparatus. Product bearing the well known trade mark EXAX and NORMAX and "K" brand will be included as well as the Kimble Blue Ring Yellow Back thermometer.

AMES COMPANY, INC.
Elkhart, Indiana

Ames Company representatives will be glad to discuss and demonstrate Clinitest, Bumintest, Acetest and Hematest, simplified test for the detection of urine, sugar, albumin, acetone bodies and occult blood.

LIPSHAW MANUFACTURING COMPANY
Detroit, Michigan

Lipshaw Manufacturing Company welcomes this opportunity to display and demonstrate our new No. 37 Rotary Microtome and the Diamond Automatic Microtome Knife Honing Machine. We will also display microtome knives, strops, hones, tissue dehydrating and embedding equipment, autopsy knives and many other items of interest to the tissue technician.

FISCHER SCIENTIFIC COMPANY
New York, N. Y.

Fischer Scientific Company, Eimer and Amend Division, will exhibit their Modern Laboratory Appliances. Included in the exhibits will be the Fischer Electrophotometer, the Fischer Electro Hemometer, Fischer Incubator, Berkeley Microtome Knife Sharpener and Tissuemat.

BALTIMORE BIOLOGICAL LABORATORY
Baltimore, Md.

B-B-L Products for the Bacteriological Laboratory will be displayed. The exhibit will include dehydrated culture media as well as completed media in special screw-capped tubes and bottles, and will feature a colorful display of interesting cultures. Various items of apparatus and equipment will be available for examinations.

A. S. ALOE COMPANY
St. Louis, Mo.

New and recent items of interest to medical technicians. Items for operation and function such as the Aloe Bath, for Prothrombin; Aloe precision Microtome Knife Sharpener; the Electro-Decalcifier for decalcification of bony tissue; the new Deeminizer for obtaining distilled water by Ionion exchange, a valuable new instrument which will determine not only hemoglobin and cell diameter but also red and white cell count.

DIFCO LABORATORIES
Detroit, Michigan

The Difco exhibits for the Medical Technologists Convention will consist of a display of clinical laboratory reagents for standardized culture media and reagents for microbiological procedures. This will include media for the detection and primary isolation of pathogenic micro-organism, together with differential media for their identification. Among these will be the media most commonly employed for culturing organisms associated with blood and respiratory and enteric infection, as well as fungus diseases.

Reagents for clinical procedures will include Bacto-Sensitivity Disks for the determination of the relative sensitivity of micro-organism to antibiotic, Bacto-penase for the inactivation of penicillin Antigen, for the Sero-diagnosis, syphillis, Bacto-Thromboplastin, Bacto-Cephalin Cholestrol Bacto-Thymol Turbidity reagent ,and Phenol-Sulponphthalein.

AMERICAN HOSPITAL SUPPLY CORPORATION
Evanston, Illinois

The American Hospital Supply Corporation will display the full line of Baxter Intravenous Solutions and Blood Transfusion equipment with all expendable accessories; complete line of highly titred, fast acting blood typing and grouping serum; Courtland Lyophilyzed Human Plasma with filter built into the bottle; reagent and standard and a selected group of Tomac specialties.

DENVER CHEMICAL MANUFACTURING COMPANY, INC.
New York, N. Y.

A new DENCO product for detection of **Albumin** will be introduced at this meeting. You are invited to visit our booth for a demonstration of this new principle in detection of albumin in urine and other body fluids. Other DENCO products on display—Galatest Powder (Sugar Test Denco) and Acetone Test Denco.

INTERNATIONAL EQUIPMENT COMPANY
Boston, Mass.

We will feature in our display our new Size 1, Model SBV, and Size 2, Model V Centrifuges. These machines incorporate a number of improvements in mechanical design which will be of interest to the users

of our equipment, and in addition have been greatly improved in appearance and performance. Also displayed will be our Clinical Model Centrifuge and special accessories for Hematocrit determinations, as well as a wide variety of interchangeable accessories for other centrifuge application.

ARTHUR H. THOMAS COMPANY
Philadelphia, Pa.

You are cordially invited to visit our exhibits in Space No. 29 where you will find the latest in laboratory apparatus and reagents.

THE TECHNICON COMPANY
New York, N. Y.

You are cordially invited to visit our exhibit in Space No. 5 where you will find the latest in Technicon Equipment.

CENTRAL SCIENTIFIC COMPANY
Chicago, Illinois

The Cenco exhibit features new apparatus for laboratories including the Cenco Stainless Steel Serology Bath, Microslide Dispenser, Thermostatic Electric Hot Plate, Plastic Agglutination Plate, Aneroid Barometer, Hydrometer, Culture Tubes with deep formed plastic cap, the Cenco Bakelite Test Tube Support and other Clinical items.

BOOKS:

Harvard University Press
Cambridge, Mass.

Appleton-Century-Crofts, Inc.
New York, N. Y.

McGraw-Hill Book Co.
New York, N. Y.

The Blakiston Co.
Philadelphia, Pa.

W. B. Saunders Co.
Philadelphia, Pa.

Charles C. Thomas Publisher
Springfield, Illinois.

JARRELL-ASH COMPANY
Boston, Massachusetts

The latest model Leitz panphot universal microscope for photomicrographic equipment will be shown operating. This is the most complete research and routine combination instrument manufactured. Other Leitz, Zeiss, American Optical and Bausch & Lomb routine and research equipment will be on display with latest light sources and specialties.

ORTHO PHARMACEUTICAL CORPORATION
Raritan, New Jersey

Ortho cordially invites you to Booth No. 13 where the complete line of Ortho diagnostic reagents will be on display. Featured will be the Ortho line of Blood Typing Sera, including Anti-Rh, Anti-Hr, Blood Grouping, and Anti-Human Serum for the Coombs test.

E. F. MAHADY COMPANY
Boston, Massachusetts

Our exhibit will consist of equipment used in bacteriological, pathological, serological and bio-chemical laboratories.

MACALASTER BICKNELL COMPANY
Cambridge, Massachusetts

Our booth will include a general array of laboratory supplies and equipment in which our services are most complete. We manufacture special research glass apparatus to specifications. We cordially invite conventionites to visit our factory in Cambridge, which is but a short distance from Swampscott. We are also purveyors of the Fenwal System for the preparation and storage and dispensing of hospital intravenous solutions, as well as whole blood and plasma and we will demonstrate briefly the most up-to-date apparatus and ideas in this regard.

C. M. Kortuem